The synthesis of 8-hydroxyellipticine and related compounds.

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THE SYNTHESIS OF 8-HYDROXYELLIPTICINE

AND RELATED COMPOUNDS

Submitted by DAVID MAXWELL DOLMAN

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the University of Bath

1982

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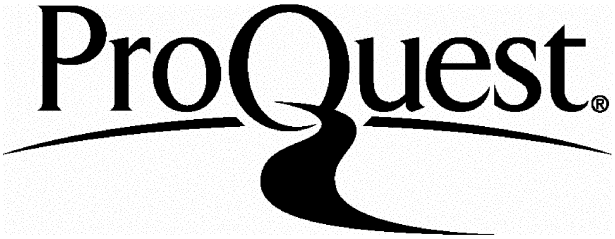


Summary

The ellipticine alkaloids or pyrido[4,3-b]carbazoles have been found to be effective against various forms of cancer, in particular leukemia, and as these compounds exhibit a low toxicity in mammals a great deal of work has been devoted to their synthesis and pharmacological testing. 9-Hydroxyellipticine has proved to be the most effective and interest in other hydroxyl derivatives naturally followed. This work is largely concerned with the synthesis of the new derivative 8-hydroxyellipticine.

Of the various known methods for preparing pyrido[4,3-b] carbazoles we chose a procedure developed at an earlier time in this laboratory. This involves a Grignard condensation between an appropriately substituted indolylmagnesium bromide and a haloethylpyridine to give the corresponding indolyl-pyridylethane. Subsequent manipulations of the pyridine ring then lead to ring closure and aromatization. This method demands the preparation of suitably protected 6-hydroxyindoles on a relatively large scale, and a great deal of labour was devoted towards the development of satisfactory routes for this purpose. Finally this was achieved, and the rest of the synthesis carried out, with appropriate modifications, to give 8-hydroxyellipticine. During the course of our work Professor J.P.Rosazza of the University of Iowa isolated 8-hydroxyellipticine in small

quantity, as an ellipticine metabolite of the micro-organism Aspergillus alliaceus. Thus, our synthesis served to provide larger quantities and to confirm the original structural assignment.

We next turned our attention to a consideration of methods for the direct electrophilic substitution of various groups into the ellipticine skeleton. This hitherto neglected area proved fruitful and 9-bromo and 9-nitro ellipticines were prepared by a much simpler and higher yielding route than any previously used.

An attempt was also made to modify and improve the potentially useful ellipticine synthesis first carried out by Stillwell[11] at Harvard University. The synthesis in its original form gave disappointingly low yields and we sought to improve this. An unexpected result of this, was the isolation of a new pyridine derivative from one of the reactions in our proposed route, but we were unable to complete this last topic due to lack of time. However, a possible method of achieving this is presented.

The thesis begins with a brief discussion of the occurrence and isolation of the ellipticines, and this is followed by a review of the various published syntheses of pyrido[4,3-b] carbazoles. In view of the recent interest in the metabolism

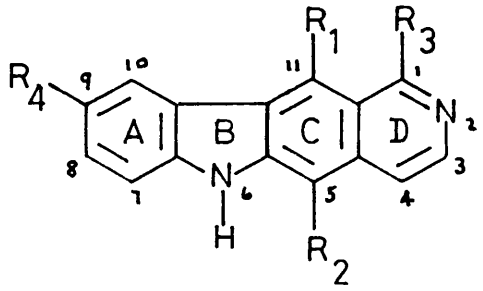
of ellipticine these experiments are also discussed. As so much work with these compounds has been aimed at achieving clinically useful derivatives this thesis concludes with a summary of the pharmacology of ellipticine derivatives.

Contents

INTRODUCTION

Occurrence and Discovery of Ellipticine

Ellipticine or 5,11-dimethyl-6H-pyrido[4,3-b]carbazole (1, $R_1 = CH_3$, $R_2 = CH_3$, $R_3 = H$, $R_4 = H$) occurs as an alkaloid in small evergreen bushes and trees of the family Apocynaceae that occurs in the tropical and sub-tropical regions of the world; typical sources being Hawaii, Fiji, South America and Australia. For example, the alkaloid was first isolated by Goodwin and his associates[1] in 1959 from the leaves of Ochrosia elliptica Labill together with several other bases including 9-methoxyellipticine (1, $R_4 = CH_3O$).



(1)

Since its initial isolation, phytochemical investigations have shown that ellipticine together with its 9-methoxy derivative occurs in plants from several other genera of the Apocynaceae, including Tabernaemontana, Peschiera and Aspidosperma[2].

Related to ellipticine is the alkaloid Olivacine (1, $R_1 = H$, $R_2 = CH_3$, $R_3 = CH_3$, $R_4 = H$) also found in plants of the Apocynaceae family[3]. Ellipticine, 9-methoxyellipticine and Olivacine all show anti-cancer activity and there is hope that this property

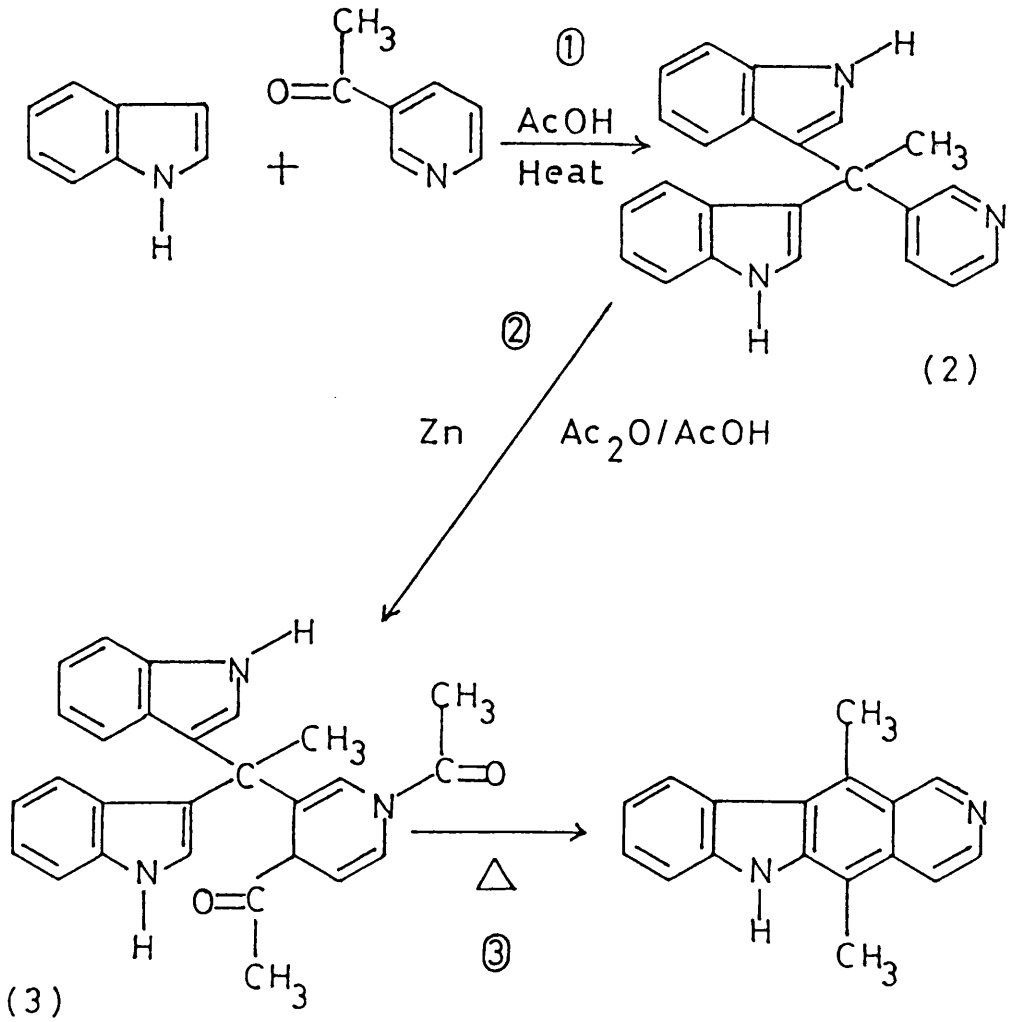
will prove useful in chemotherapy because these bases have shown relatively low mammalian toxicity. Since the natural products are insoluble in water, interest has focussed upon derivatives with polar substituents, in particular the hydroxyellipticines.

9-Hydroxyellipticine obtained by demethylation of the 9-methoxy compound, has far greater activity than the natural alkaloid itself, and Lallemand[56] has found that in the rat, ellipticine is hydroxylated at C-7 and C-9. Recent work by Rosazza[63], has also revealed that 8-hydroxyellipticine is formed together with some of its 9-hydroxy isomer when ellipticine is fed to the micro-organism Aspergillus alliaceus, and it is intriguing that the two biological systems should metabolise the alkaloid in different ways, (see p.64 for more details of these experiments).

Synthetic methods employed for Ellipticine and its derivatives

The first synthesis of ellipticine (see Scheme 1) was achieved by Woodward and his group[4] in the same year as the alkaloids isolation. This synthesis was remarkably short and direct, but unfortunately it gave a very low overall yield. Thus, while it provided a useful proof of structure it could not be considered as a route of general applicability.

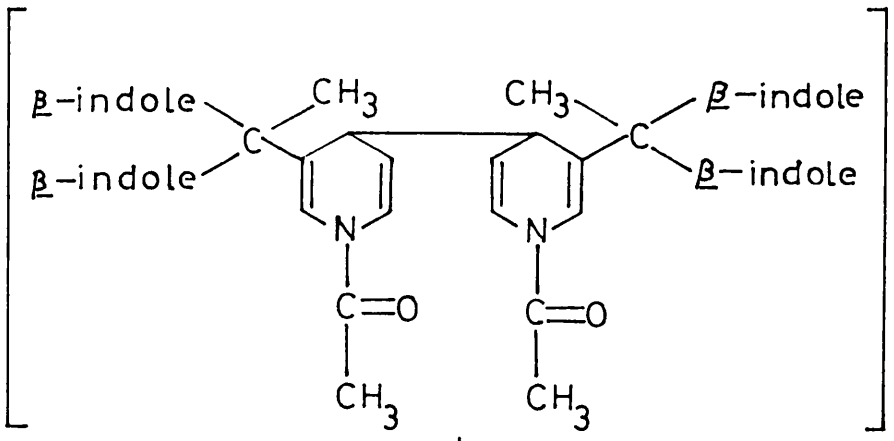
Scheme 1.



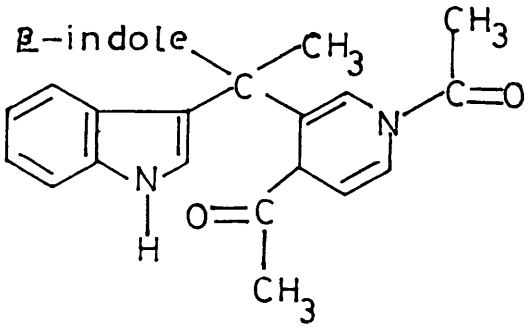
Indole was condensed with 3-acetylpyridine in hot acetic acid to give 1,1-bis (3-indolyl)-1-(3-pyridyl)ethane (2), this was subjected to the Wibaut-Arens[5] reductive acetylation reaction using zinc and acetic acid/acetic annhydride, to give the derivative (3). The final oxidative ring closure was achieved by the use of severe pyrolytic conditions, followed by sublimation, and the yield in this last stage was only 2%. Significantly, the yields of the other individual stages were not quoted, but it is likely that the productivity of stage 2 (Scheme 1.), was low.

The initial reaction between the indole and the acetylpyridine proceeds so that two molecules of the indole combine with one of the pyridine, unless either of the reactants have a bulky substituent; so orientated as to preclude the formation of compounds of type (2). Some reactions in the latter class have been found[6] to possess interesting synthetic applications (see discussion section page 126).

Unfortunately the 2:1 product is a poor substrate for the next stage of the synthesis, since the Wibaut-Arens reductive acetylation of the pyridine (2) proceeds through a complex dimeric species (4) which disproportionates into starting material and the 1,4-diacetyl-1,4-dihydropyridine (3).

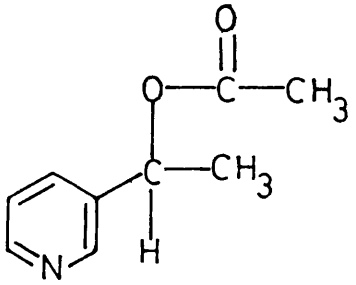


(4)

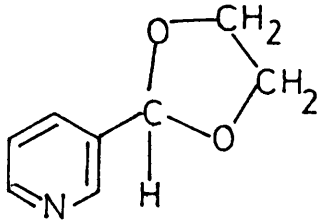


(3)

The formation of the postulated intermediate(4) is thus very sensitive to steric effects and it has been shown in this laboratory[7] that its ease of formation is much reduced when the pyridyl units contain bulky substituents. For example, the 3-substituted pyridines (5) and (6) do not react under the Wibaut-Arens conditions



(5)

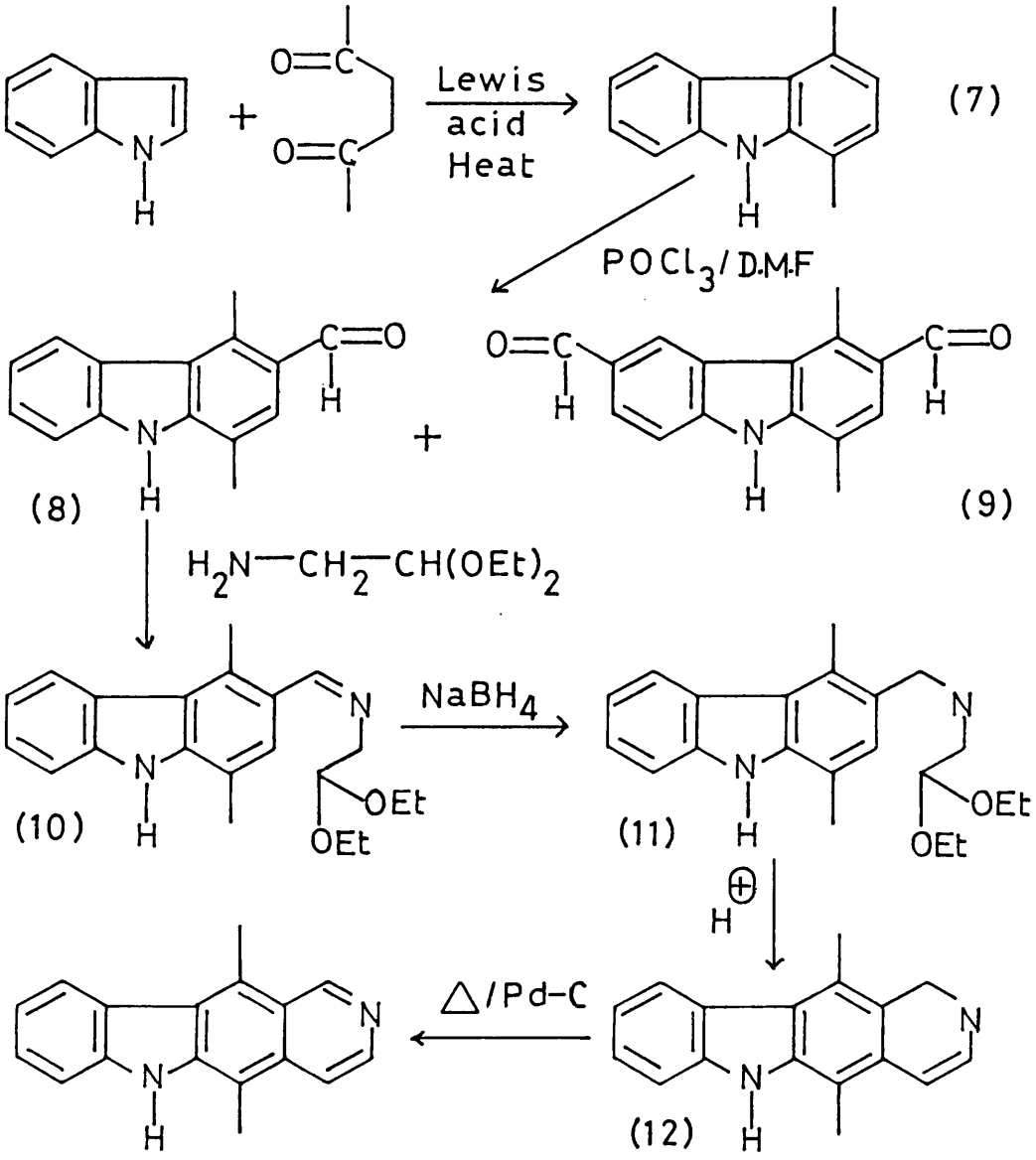


(6)

It is clear that in the normal 2:1 reaction, where intermediates such as (4) are formed these steric problems are rendered extreme, and impose a restriction on the types of substituents the pyridine may contain. This further detracts from the usefulness of this procedure.

A second synthesis was published in 1962 by Cranwell and Saxton[8]. These workers prepared 1,4-dimethylcarbazole (7) in an optimised yield of 36% by the condensation of indole with 2,5-hexadione in the presence of a Lewis acid catalyst. Vilsmeier formylation of (7) under controlled conditions gave the formyl derivative (8), together with some 3,6-disubstituted product (9). These were separated, and the 3-substituted product (8) was condensed with 2,2-diethoxyethylamine to afford the Schiffs base (10) in good yield. However, all attempts to cyclise this compound failed, and the authors found it necessary to reduce the imine (10) to the amine (11) and ring close this product under acid conditions to give the dihydroellipticine (12). It was then necessary to dehydrogenate the 1,2-dihydroellipticine (12) to the fully aromatic tetracycle by heating with palladium carbon. This treatment gave ellipticine, but in a disappointingly low yield of 7%. This route summarized in(Scheme 2)has potential practical utility due to the small number of steps and the availability of the starting materials, but the final stages are not very efficient, which reduces its value as a general synthesis.

Scheme 2

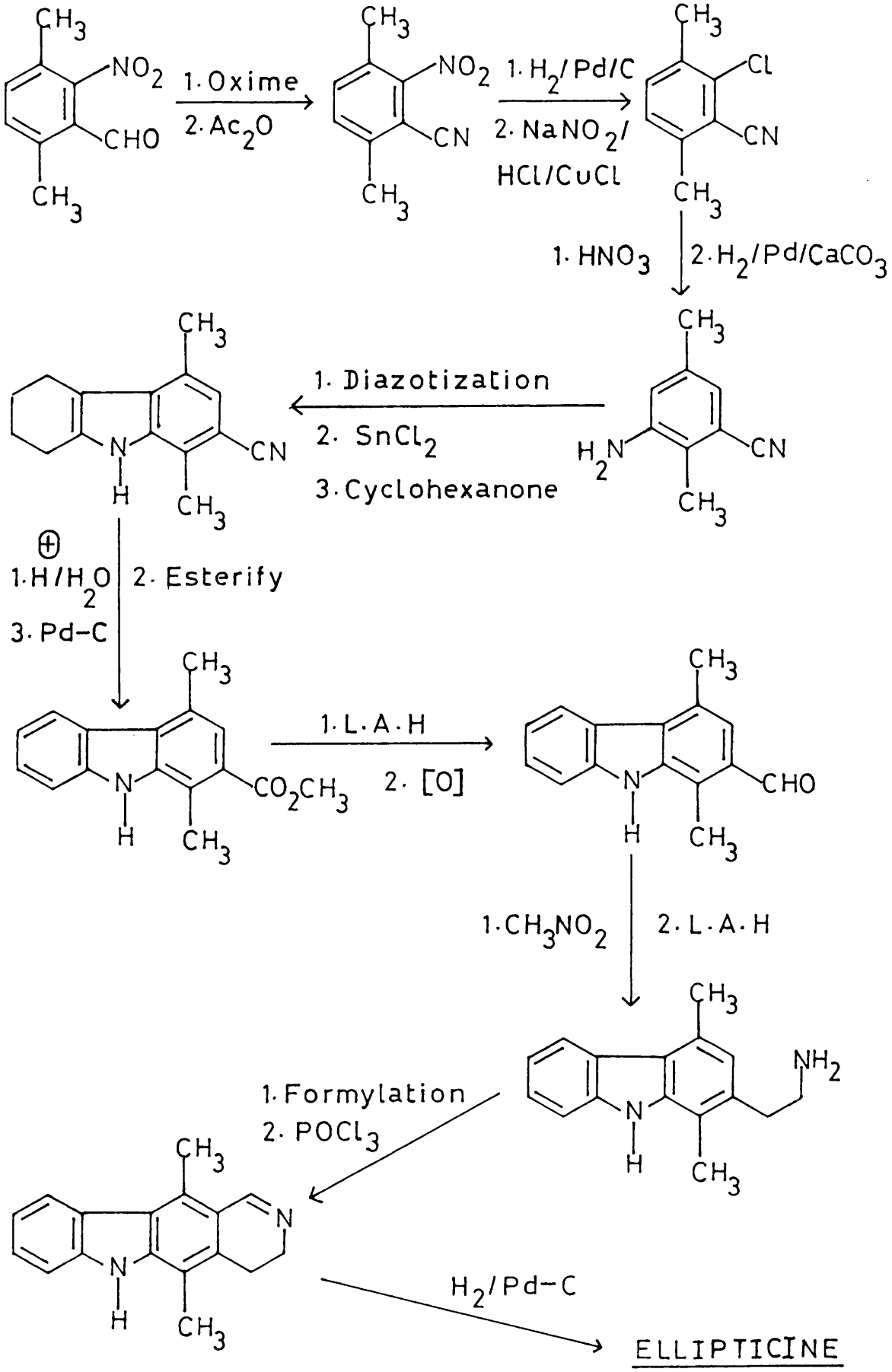


Two more syntheses soon appeared, but neither method has practical value. The first by Govindachari[9] and his associates in 1963 took the novel approach of starting with a pre-formed ring C system. The route (outlined in Scheme 3) represents a fine example of the techniques of classical organic chemistry, but the large number of steps required to construct the tetracyclic system are obviously disadvantages in a general preparative route, although Mosher[10] used a modified sequence to prepare the related alkaloid olivacine.

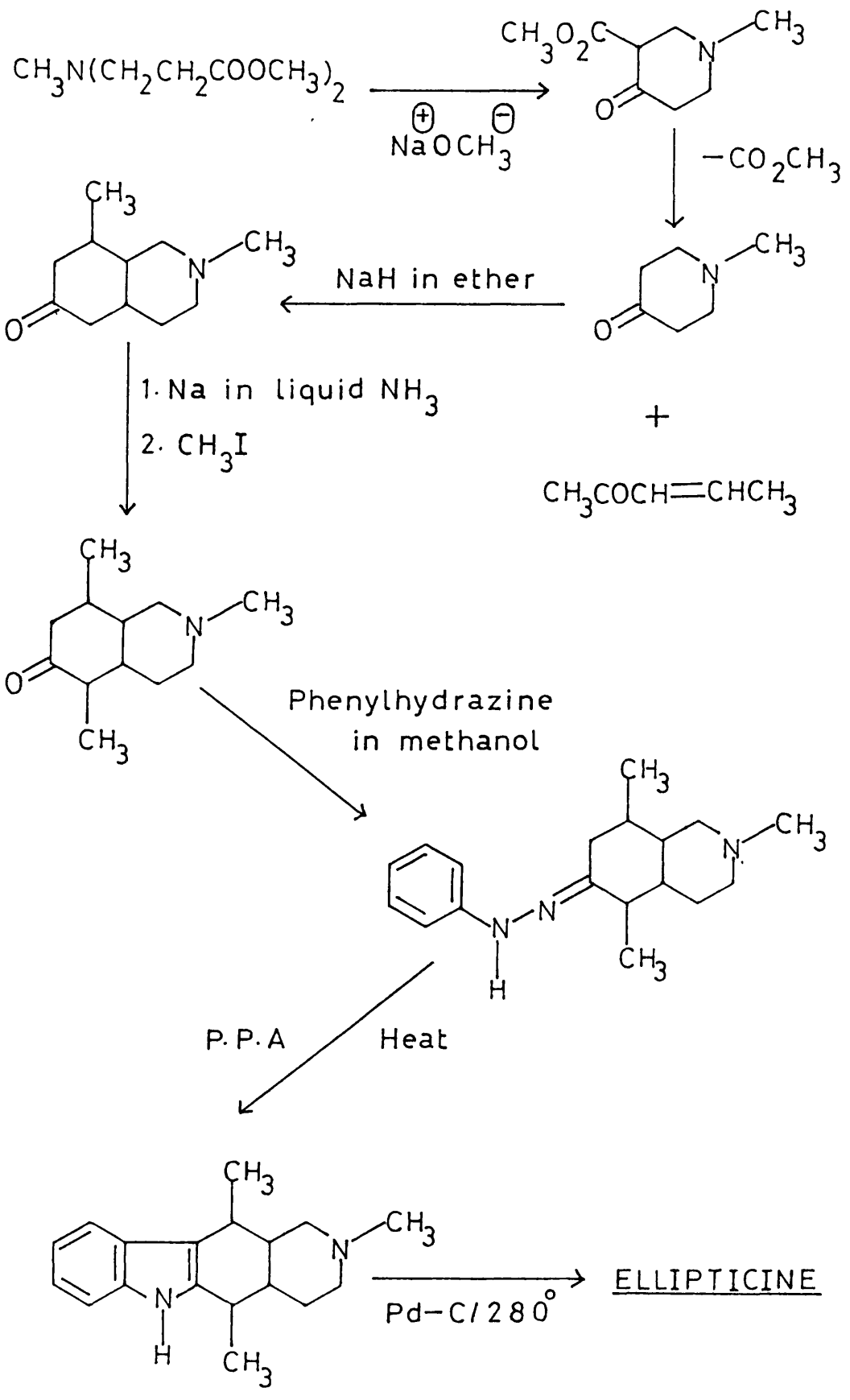
The second synthesis by Stillwell[11] is also unusual, because it involves the prior formation of rings C and D from aliphatic precursors, followed by a Fischer-indolisation reaction to form the rest of the tetracyclic system. High temperature dehydrogenation of the resulting octahydroellipticine completes the synthesis. This route was based on the often frustrating premise of satisfactory model reactions, but unfortunately in the actual synthesis yields were poor; only sufficient product was obtained to allow its characterisation by physical methods. This approach is outlined in (Scheme 4).

A point to note here is that the final high temperature dehydrogenation step constitutes a severe limitation upon the choice of ring substituents that can be used successfully in this synthesis, as thermally labile groups, or those that are easily oxidized such as amino functions, will cause degradation of the ring system to intractable polymeric materials. Such a constraint also applies to some of the more modern approaches to ellipticines.

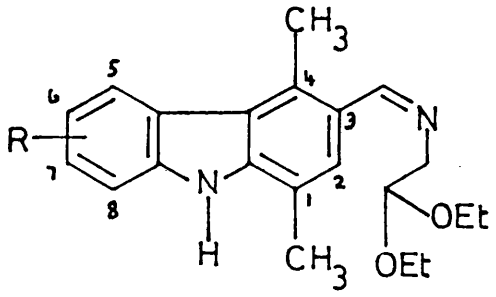
Scheme 3.


1. Oxime
2. Ac_2O
1. H_2/Pd/C
2. $NaNO_2$/ HCl/CuCl
1. HNO_3
2. H_2/Pd/$CaCO_3$
1. Diazotization
2. $SnCl_2$
3. Cyclohexanone
1. $H^{\oplus}$/H_2O
2. Esterify
3. Pd-C
1. L.A.H
2. [O]
1. CH_3NO_2
2. L.A.H
1. Formylation
2. $POCl_3$
H_2/Pd-C
ELLIPTICINE

Scheme 4.


CH3N(CH2CH2COOCH3)2
NaOCH3
CH3O2C
CH3
N
O
−CO2CH3
NaH in ether
1. Na in liquid NH3
2. CH3I
+
CH3COCH=CHCH3
Phenylhydrazine
in methanol
P.P.A
Heat
H
Pd−C/280°
ELLIPTICINE

The full potential of the original work by Cranwell and Saxton was realised in 1967 by Dalton[12] and his team in Australia, who used the route to make several derivatives of ellipticine. This group found that the concentration of acid, used to cyclize Schiffs bases of the type (13) is critical.



(13)

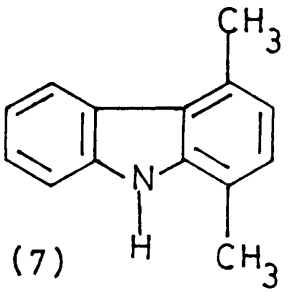
Employing 95-100% ortho-phosphoric acid to effect the Pomeranz-Fritsch ring closure they found that cyclization of the imine (10) and substituted imines of type (13) occurred directly, to give ellipticines, thus obviating the two final steps of the original approach. The yields also, were very much improved; being ~30% overall; where R = H, and better for some derivatives (see Table 1 below).

During the course of this work a number of interesting facts emerged. Firstly, it was found that electron donating groups in the C-6 position of carbazole intermediates of the type (13) assisted the final ring closure, whereas electron withdrawing functions at C-6 severely limited, and in some cases inhibited ring closure.

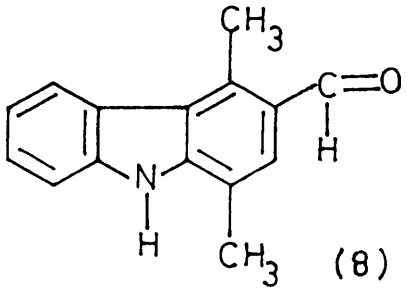
Table 1

Substituent at C-6	Yield of ellipticine
H	33
CH_3	50
OCH_3	56
Br	45
NO_2	11

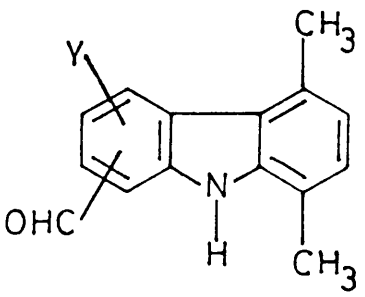
Secondly carbazoles of type (7) with electron donating substituents in the non-methylated ring, were found to give rise, on Vilsmeier formylation, to a mixture of the required 3-formyl compound (8) and compounds of type (14) where the substituent function has entered the ring bearing the activating group.



(7)



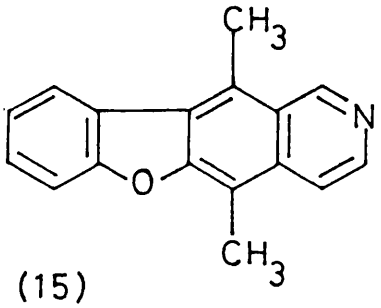
(8)



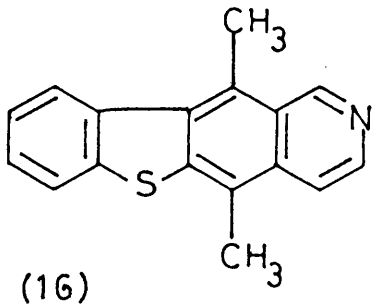
(14)

Where Y is an electron releasing substituent.

The harsh conditions of the final step, involving the use of polyphosphoric acid (P P.A) at a temperature of 160-180^{o} are undesirable where acid sensitive or labile substituents are required. Despite these limitations this route represents the first practical approach, and in addition to ellipticines, it has been employed by Elmes and Swan[13] to synthesize the two related tetracycles (15) and (16).



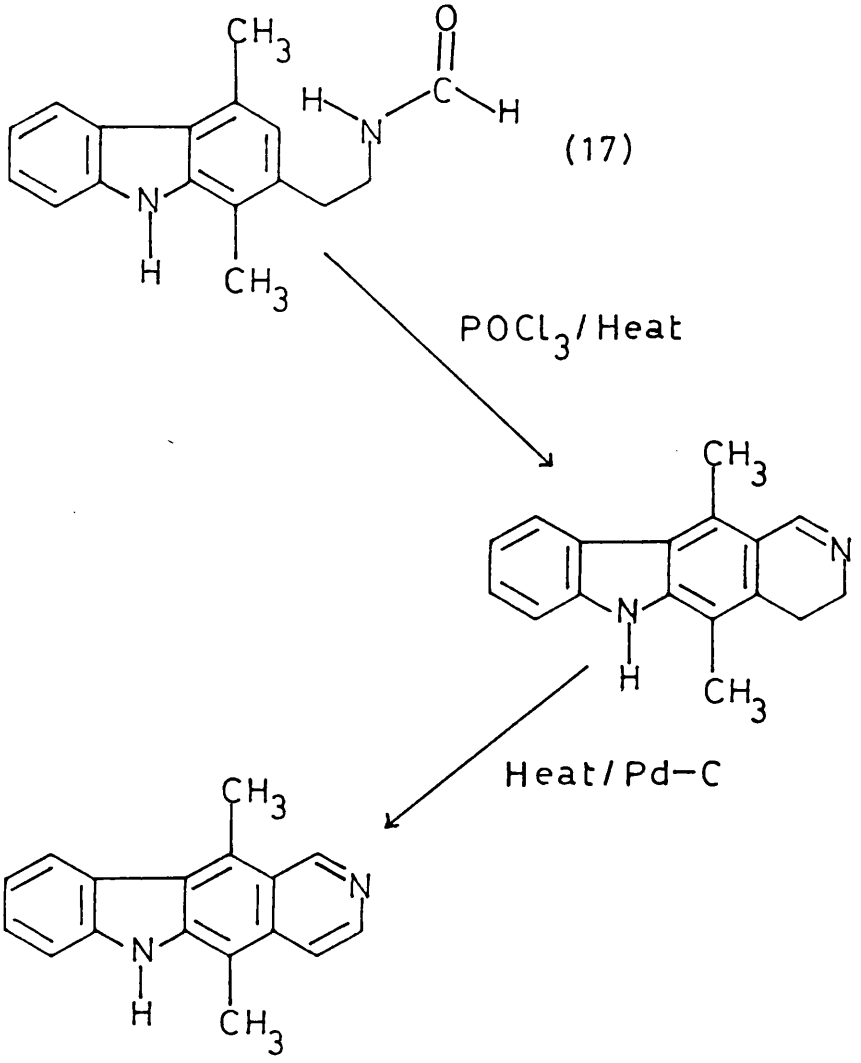
(15)



(16)

Thus, 5,11-dimethylbenzofuro[2,3-g]isoquinoline (15) and 5,11-dimethylbenzothieno[2,3-g]isoquinoline (16) were synthesized from 1,4-dimethyldibenzofuran and 1,4-dimethyldibenzothiophen respectively. Unfortunately both of these compounds are inactive against experimental tumour systems.

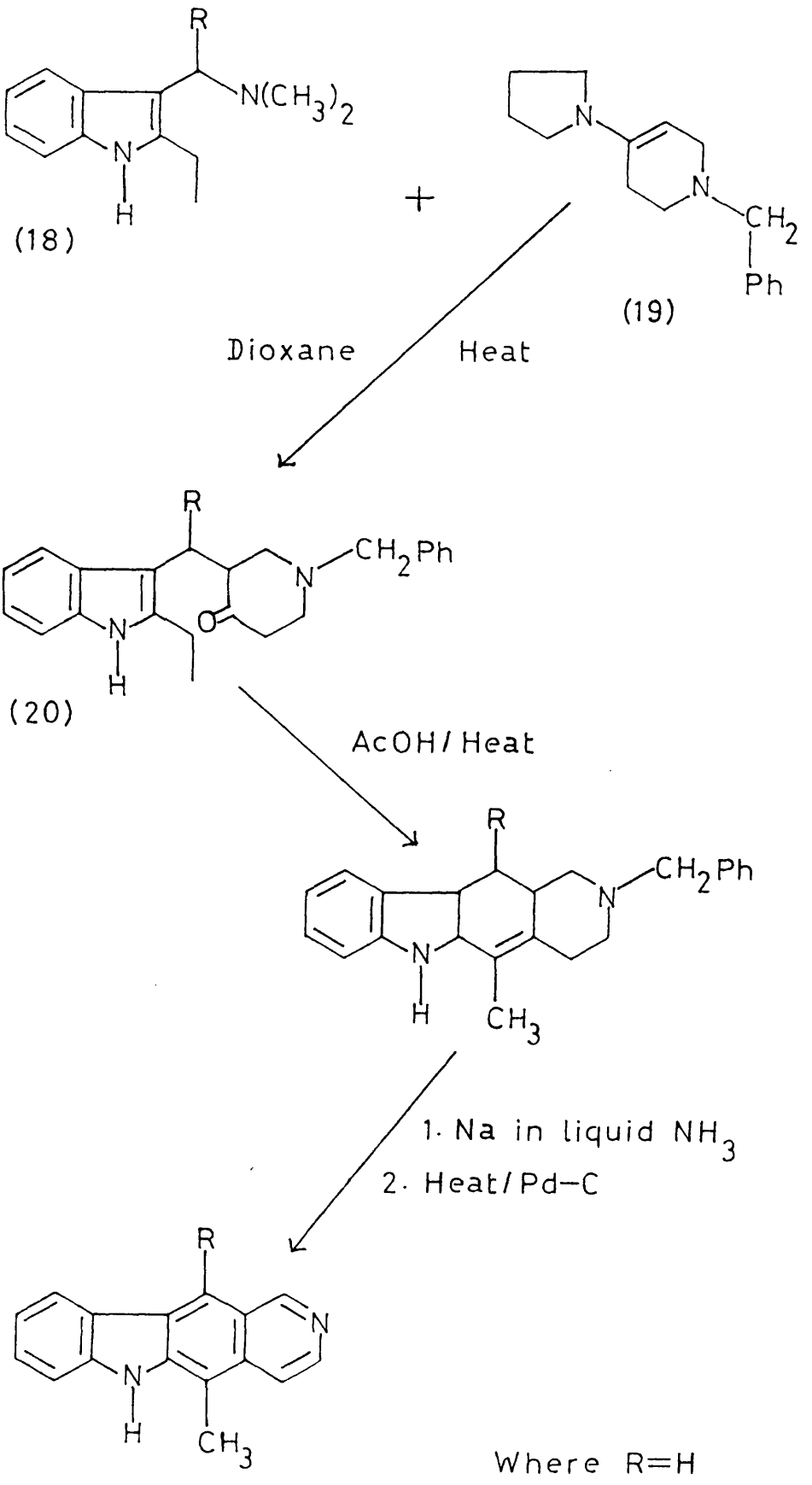
A modification of this synthesis has been patented[14] by a Swiss group. These workers prepared the 2-substituted carbazole (17) and achieved the cyclization using phosphorus oxychloride. This furnished 3,4-dihydroellipticine, which was subsequently dehydrogenated to the alkaloid.



French chemists have also expended a great deal of effort in this field and Goffic and his associates[15] have pioneered two syntheses of the ellipticine skeleton. The first is shown in (Scheme 5).

Reaction of 2-ethylgramine (18) with the enamine (19) in anhydrous dioxane followed by hydrolysis gave the piperidone structure (20). This was cyclized to the tetracyclic skeleton by heating in acetic acid. Debenzylation and dehydrogenation in boiling decalin gave 11-demethylellipticine.

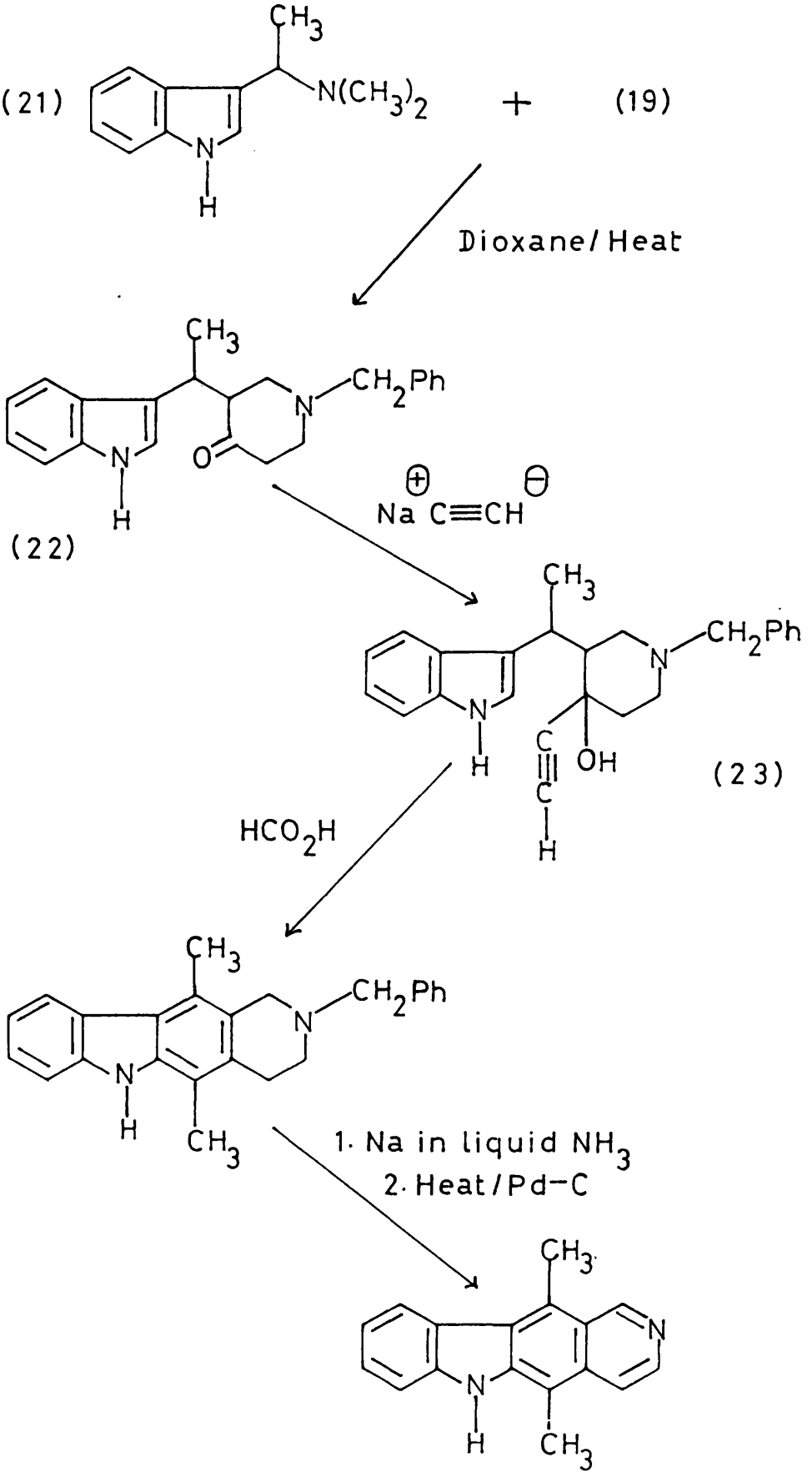
Scheme 5.


R
N(CH3)2
N
H
(18)
+
N
N
CH2
Ph
(19)
Dioxane
Heat
R
CH2Ph
N
O
N
H
(20)
AcOH/ Heat
R
CH2Ph
N
N
H
CH3
1. Na in liquid NH3
2. Heat/Pd–C
R
N
N
H
CH3
Where R=H

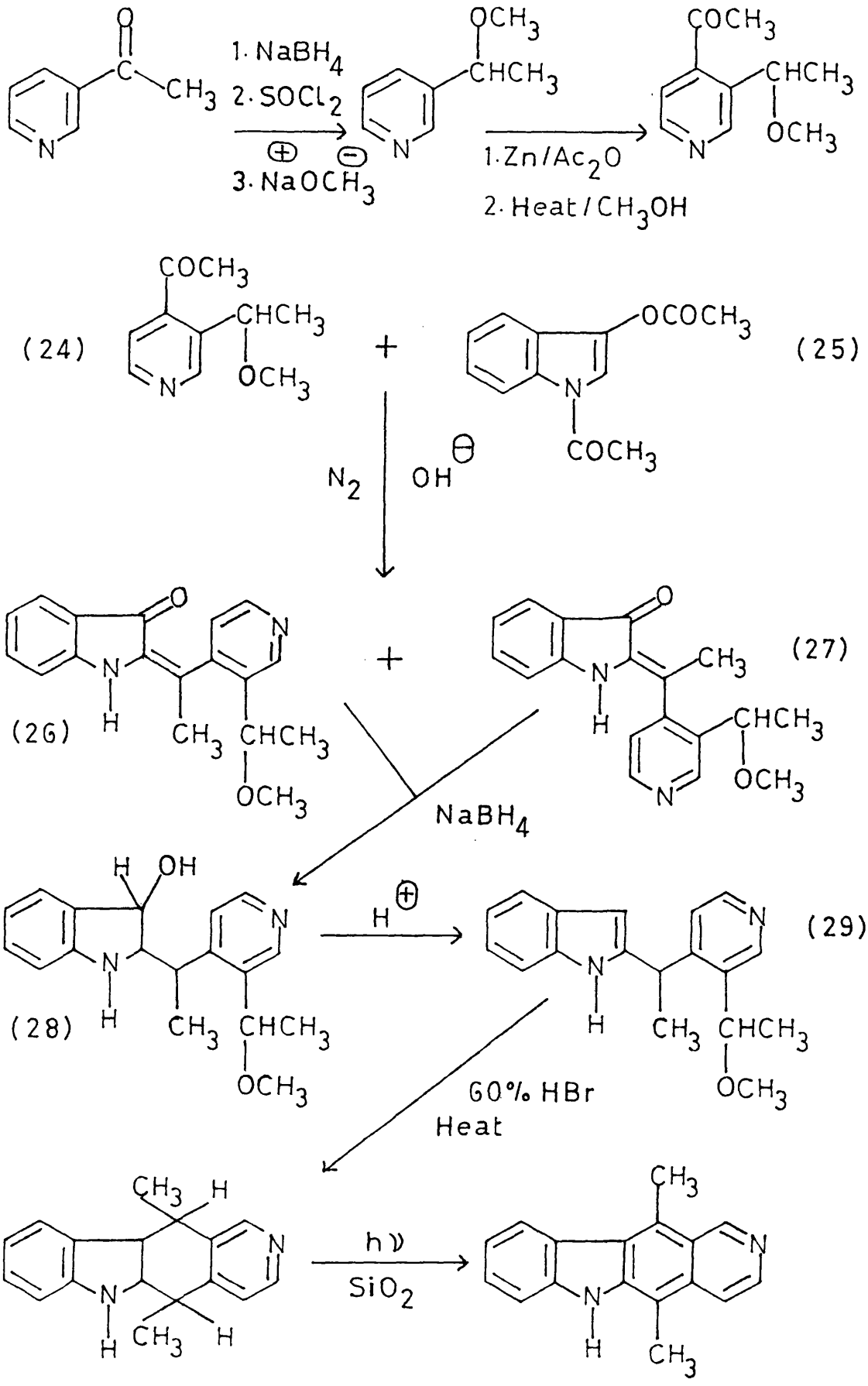
The second approach is outlined in (Scheme 6). The gramine derivative (21) was condensed with the enamine (19), as before, to yield the piperidone (22) in good yield. This was reacted with sodium acetylide to form the acetylenic alcohol (23). Treatment with formic acid gave 2-benzyl-1,2,3,4-tetrahydroellipticine which was subsequently dehydrogenated and de-N-benzylated to afford ellipticine. The overall yield from the indole was 24%.

The same year saw another new, and potentially versatile approach to the tetracyclic system, developed through work in this laboratory[16] and is outlined in (Scheme 7).

Scheme 6.

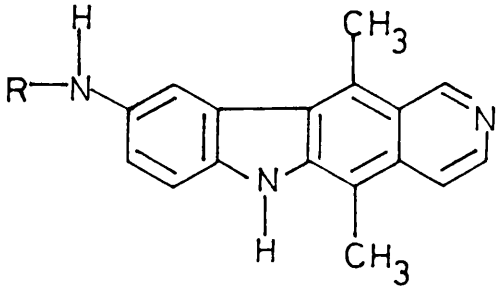

(21)
(19)
Dioxane/Heat
(22)
Na C≡CH
(23)
HCO2H
1. Na in liquid NH3
2. Heat/Pd–C

Scheme 7.


1. NaBH4
2. SOCl2
3. NaOCH3
1. Zn/Ac2O
2. Heat/CH3OH
(24)
(25)
N2
OH⊖
(26)
(27)
NaBH4
(28)
H⊕
(29)
60% HBr
Heat
hν
SiO2

The 4-acetylpyridine (24) was prepared from 3-(1-methoxyethyl) pyridine by the Wibaut-Arens reductive acetylation reaction, followed by aromatization of the product. Condensation of the masked indoxyl (25) in alkaline conditions, under a protective nitrogen atmosphere proceeded smoothly to give a mixture of the isomeric E- and Z-2-ethylidene indolin-3-ones (26 and 27) in 75% yield. Reduction of the mixed isomers with sodium boronhydride afforded the alconol (28) which denydrated under acid conditions to give the indole (29). The indole (29) was heated under reflux with 60% aqueous hydrobromic acid, and upon basification and extraction, 5,11-dinydroellipticine was obtained. Exposure of this to ultraviolet light on a silica support lead to aromatization and ellipticine itself. The overall yield of ellipticine was 31%.

Having achieved a successful synthesis of ellipticine the interest turned to the preparation of new derivatives for pharmacological testing purposes, and with this end in view modifications of (Scheme 7) were employed to prepare 9-amino, 9-phenyl and 9-bromoellipticines[17,18]. It was thought that 9-amino-ellipticine should provide a large number of other tetracycles through simple interchanges involving the amino group. This was achieved by the author, with the preparation of 9-tosylamido and 9-benzenesulphonamido ellipticines[19] (30 and 31) from the parent compound (32).

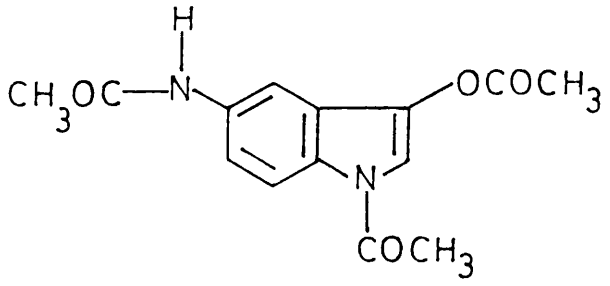


30. R = $CH_3.C_6H_4SO_2$

31. R = $C_6H_5.SO_2$

32. R = H

The method used to synthesize 9-aminoellipticine was identical to that shown in (Scheme 7) above, except that the appropriate masked indoxyl was employed. In this case, it proved necessary to use 9-acetamido-3-acetoxy-1-acetylindole (33), rather than the corresponding free amino compound, because the latter is too oxidation sensitive to allow the necessary manipulations.



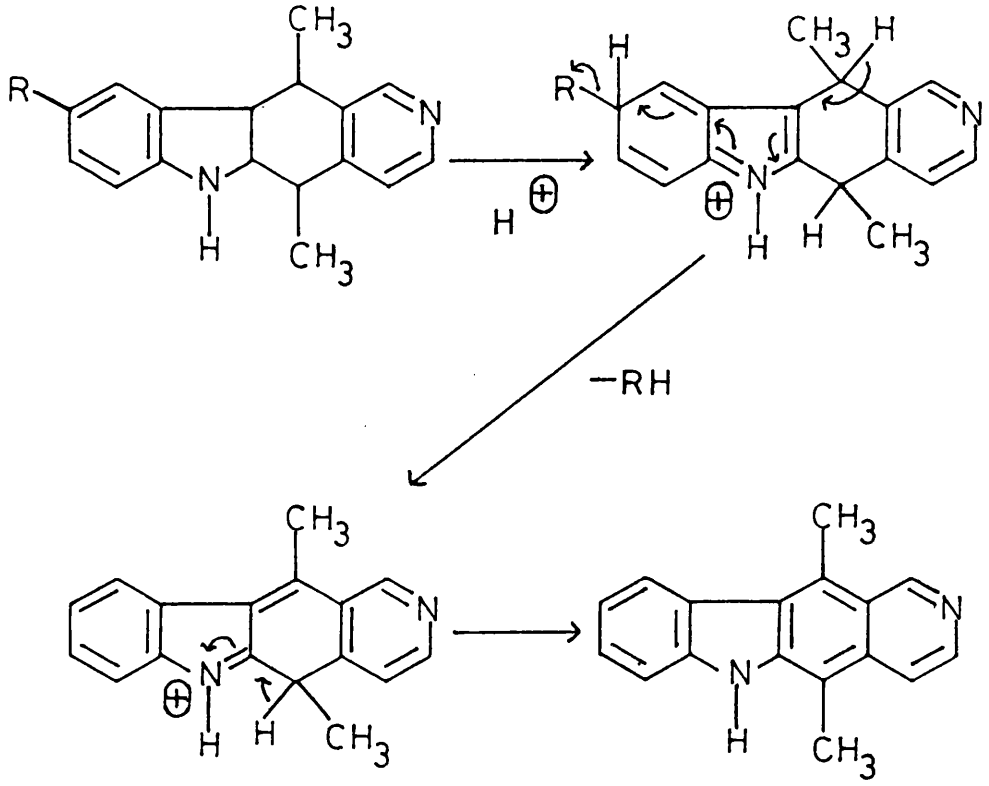
(33)

A point to note in this case was that cyclization with hot hydrobromic acid of the acetamido indole corresponding to (29, Scheme 7) proceeded directly to the fully aromatic tetracycle, thus obviating the final step involving irradiation on silica.

However, this synthesis suffered from a number of disadvantages. Firstly the reductive acetylation[5] of the pyridine component and its subsequent aromatization proceeds through a number of complex intermediates not shown above (see p. 5), and the overall yield is poor. Secondly the ring closure conditions in the final step of the ellipticine synthesis involving aqueous 60% hydrobromic acid at reflux temperature for a number of hours, are harsh, and a certain amount of ellipticine always accompanied the desired product when the indoles of type (29) bore a 5-acetamido or a 5-bromo substituent. This interesting

reaction is thought[18] to be associated with in situ oxidation of the first formed 5,11-dinydroellipticine, possibly as shown in (Scheme 8).

Scheme 8.

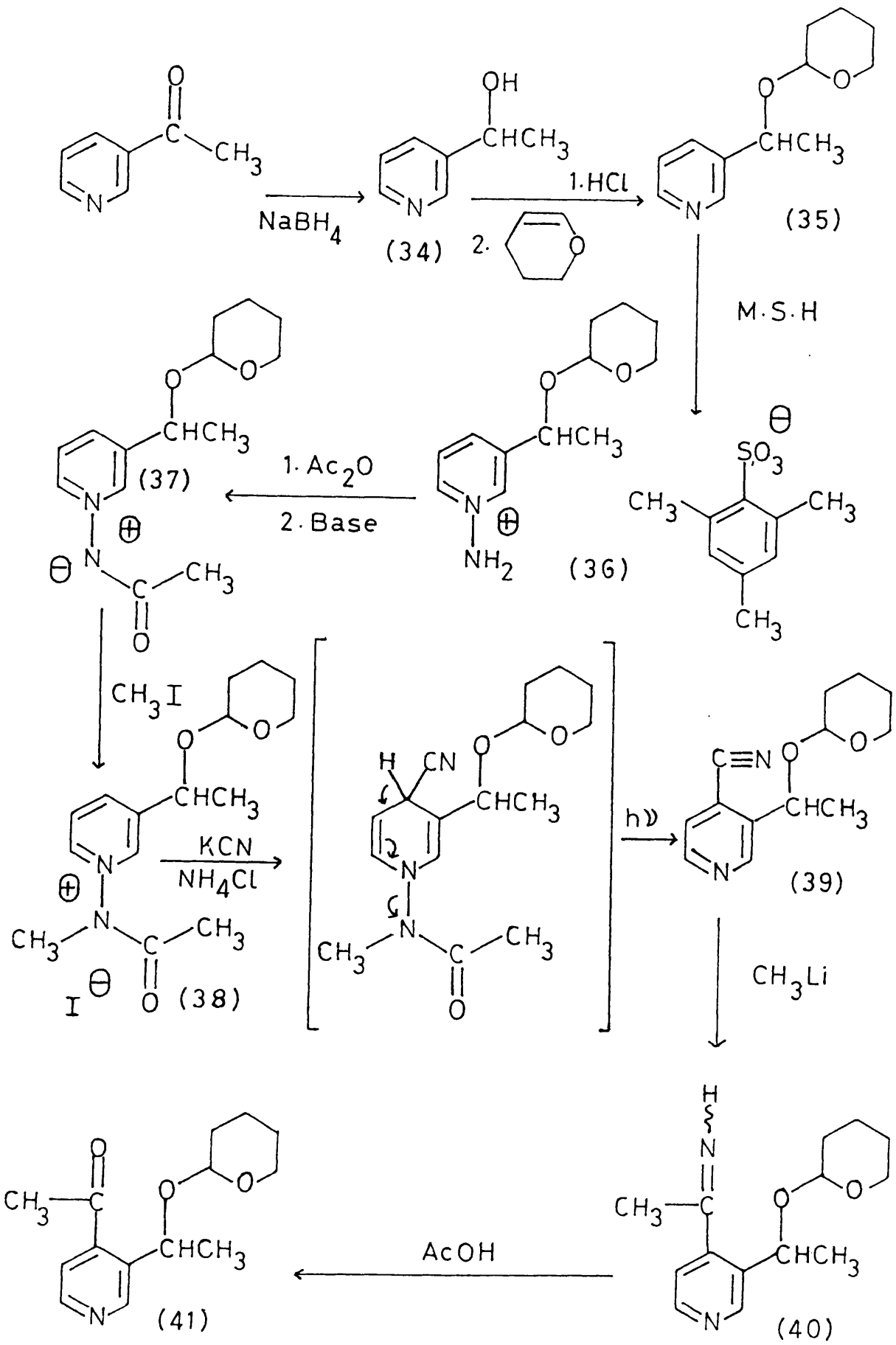


Further work in this laboratory has established that a similar loss of the 9-substituent occurs whenever that unit may form a stable anion, thus diminishing the usefulness of this approach to the synthesis of substituted ellipticines.

In view of the limitations described above it was decided[18] to replace the pyridine ether (24) in (Scheme 7) by a more readily accessible intermediate, which on being incorporated into the two position of a suitable indole derivative would require less vigorous cyclization conditions. After a considerable amount of development work the protected pyridine alconol (41) was selected, and this was synthesized as shown in (Scheme 9).

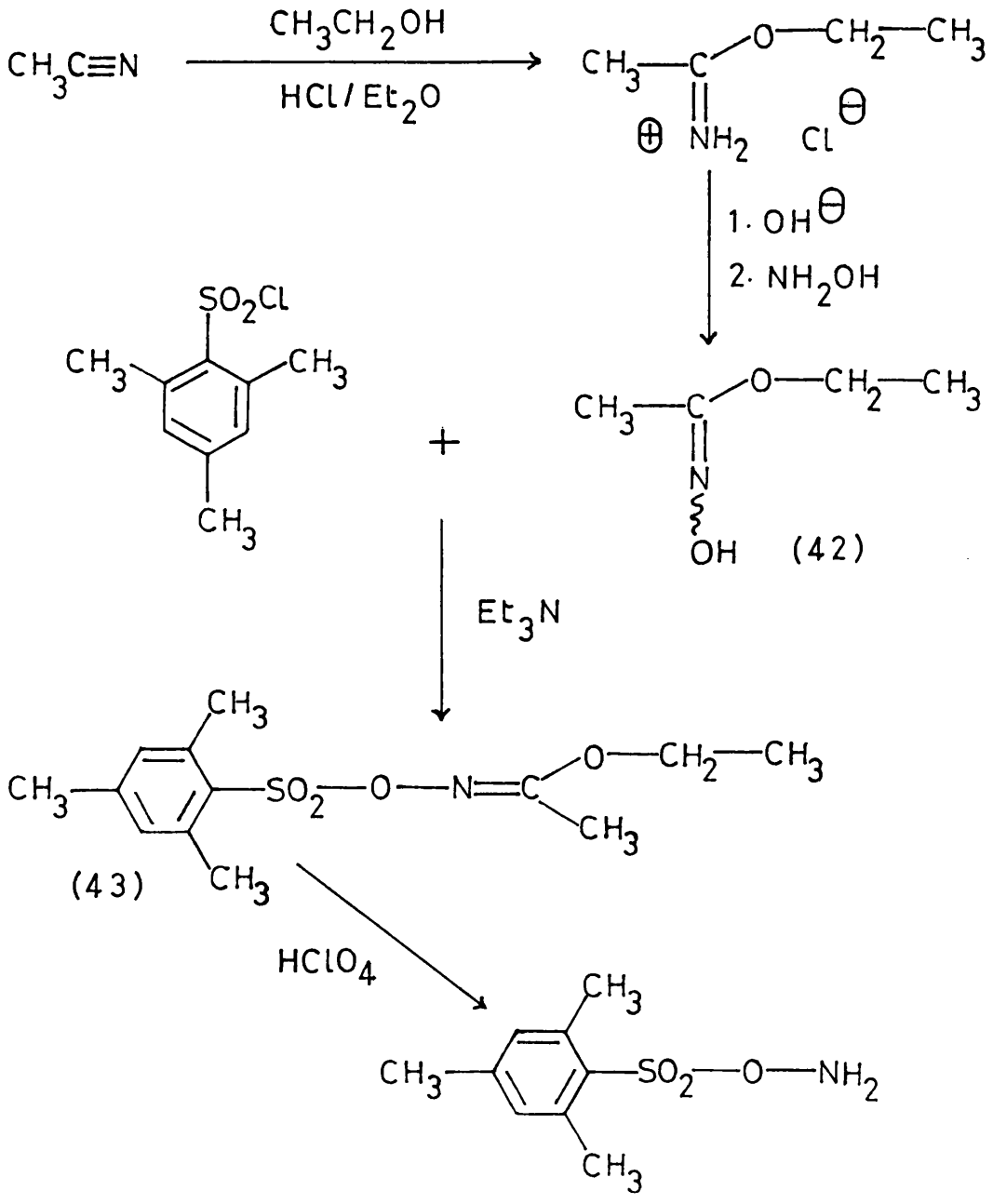
The key reaction in this sequence is the activation of the pyridine nucleus towards nucleophilic attack by cyanide ion. Hence, the pyranyloxy derivative (35) was treated with mesitylene sulphonyl hydroxylamine (M.S.H.)[20], prepared as shown in (Scheme 10). This reagent aminates the pyridine nitrogen atom to form a pyridinium salt (36), which on treatment with acetic annhydride at 0^{o}, followed by basification, afforded the Zwitter ion (37). Subsequent methylation using methyl iodide yielded the methiodide salt (38). This compound has the γ-position of the pyridine ring activated towards nucleophilic attack and the group is also sufficiently bulky to sterically innhibit reaction at the two α-positions[19,20,21]. Nucleophilic attack by cyanide ion on this compound under conditions of controlled pH gave a 1,4-dihydro-pyridine intermediate which aromatized, with expulsion of N-methylacetamide, on exposure to 'soft' ultraviolet light. Treatment of the 4-cyano compound (39) with methyl lithium produced the methylimine (40), and this was hydrolysed with glacial acetic acid at room temperature to give the required 4-acetylpyridine (41). The overall yield was 65% based on the

Scheme 9.


NaBH4
(34)
1.HCl
2.
(35)
M.S.H
(36)
1.Ac2O
2.Base
(37)
CH3I
(38)
KCN
NH4Cl
hν
(39)
CH3Li
(40)
AcOH
(41)

alconol (34). This represented a tremendous improvement with respect to the Wibaut-Arens reductive acetylation reaction previously cited (p. 4). Further developments based on this procedure gave rise to a new route to ellipticines of great potential versatility which is described in detail on (p. 62), as it is a key reaction sequence in the work that follows later in this thesis.

Scheme 10.

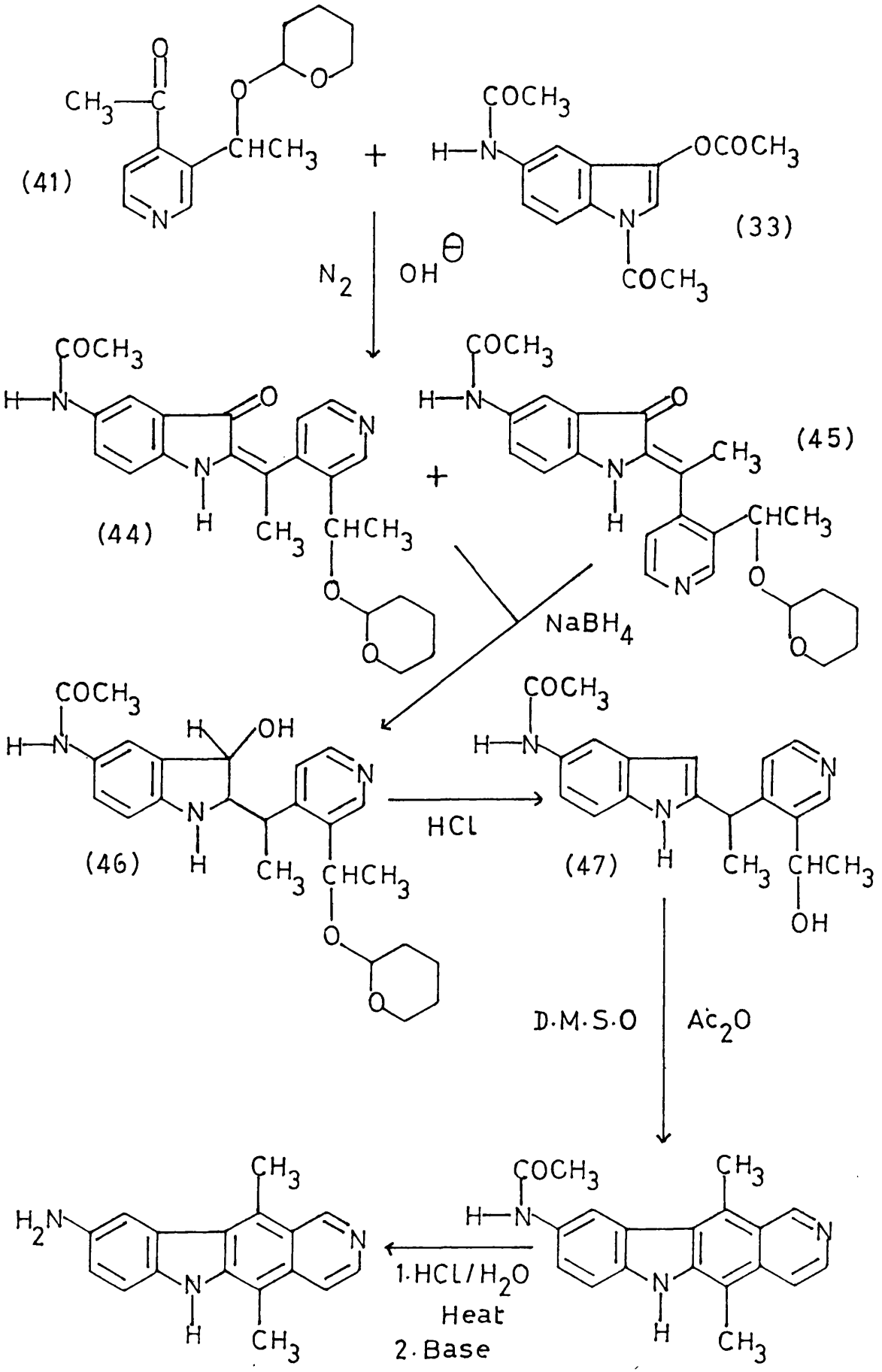


1-Ethyl-1-oximidoethane (42)was condensed with mesitylene sulphonyl chloride to give ethyl-O-mesitylene-sulphonylaceto-hydroxamate (43), which was hydrolysed with 70% perchloric acid to give mesitylene sulphonyl hydroxylamine (M.S.H.).

Having obtained the pyridine (41), it was condensed, as before, with 5-acetamido-3-acetoxy-1-acetylindole (33), under alkaline conditions as outlined in (Scheme 11).

The condensation gave the mixed indoxylidines (44) and (45) in 90% yield. These were reduced without separation to the indolinol (46) using sodium borohydride in hot ethanol. The indolinol (46) was treated with dry hydrogen chloride gas in anhydrous methanol to effect dehydration of the indolinol moiety with simultaneous hydrolysis of the pyranyloxy protecting group, thus affording the alcohol (47). This alcohol was treated with dimethylsulphoxide[22] and acetic anhydride at room temperature which caused oxidation and concomitant cyclisation to 9-acetamidoellipticine. This product was subsequently hydrolysed to the amino compound by hot aqueous hydrochloric acid.

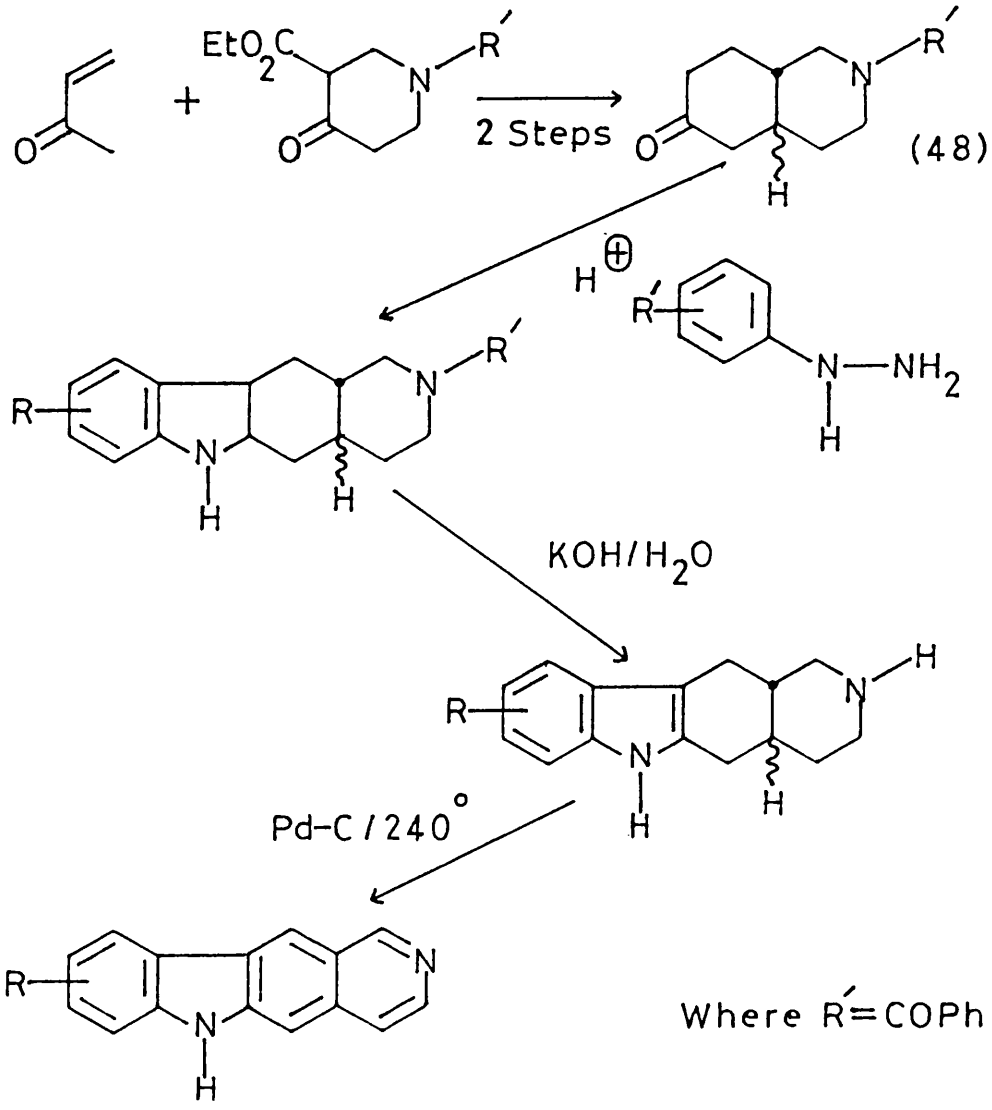
Scheme 11.


(41)
(33)
N_2
$OH^{\ominus}$
(44)
(45)
$NaBH_4$
(46)
HCl
(47)
D.M.S.O
Ac_2O
1.HCl/H_2O
Heat
2.Base

The following years saw the publication of many more synthetic routes to the ellipticines by chemists from all over the world.

A modification of Stillwell's work (Scheme 4), by Rastogi and his group[23] in India, was claimed to give rather better yields of some derivatives, but none were actually quoted so the potential of this modified approach remains unclear. These workers prepared 7-fluoro, 9-fluoro, 7-methoxy and 9-methyl-6H-pyrido[4,3-b]carbazoles by the route outlined in (Scheme 12).

Scheme 12.

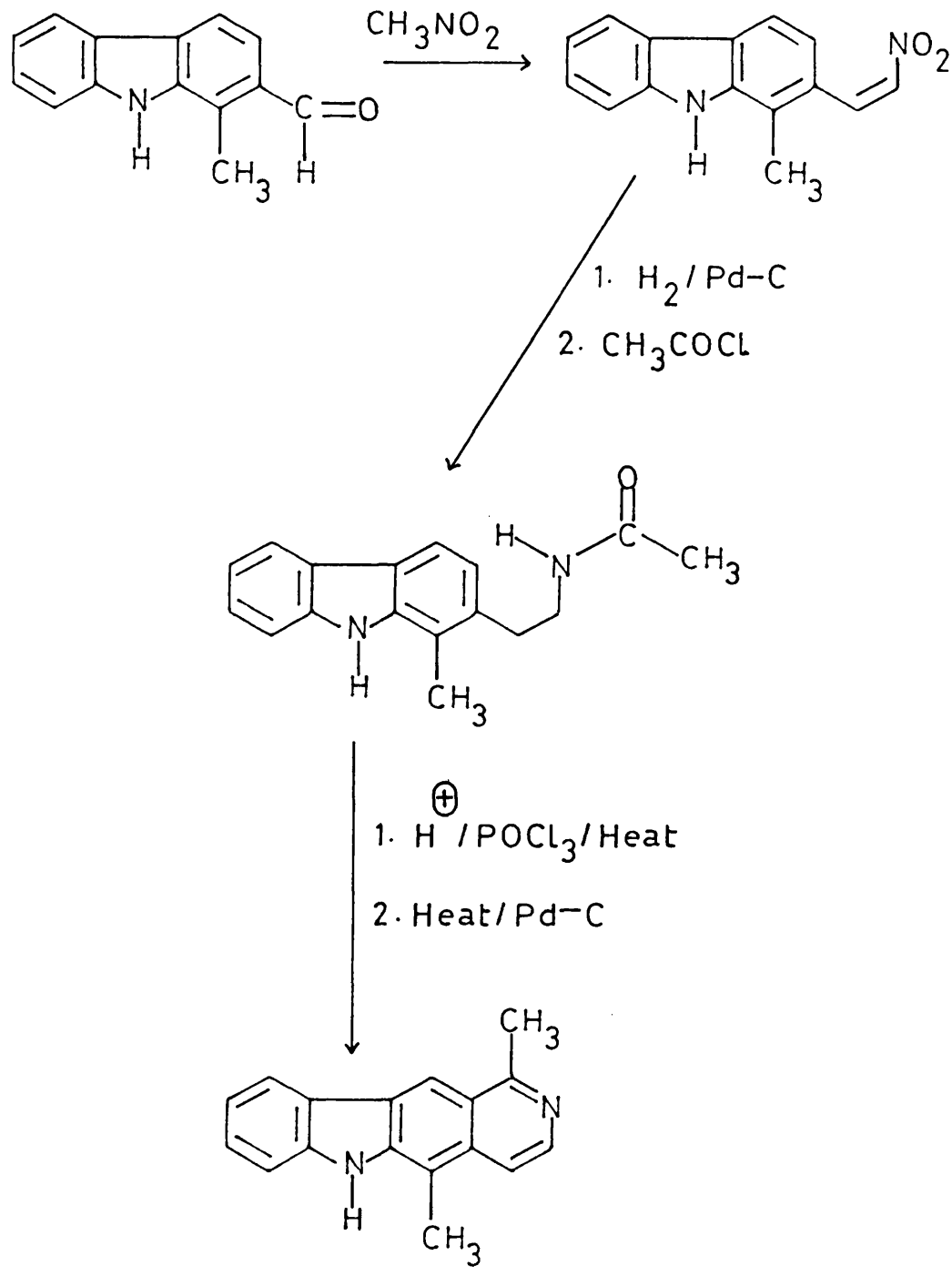


The key intermediate 2-benzoyl 1,3,4,4a,5,7,8,8a-octahydro-6(2H)-isoquinoline was prepared from 1-benzoyl-3-carboethoxy-4-piperidone and methylvinyl ketone. This product (48) was condensed with the appropriate arylhydrazine and subjected to the conditions of the Fischer indole synthesis. Hydrolysis and dehydrogenation of the product afforded the desired pyrido [4,3-b]carbazoles. Once again the high temperature dehydrogenation step is very undesirable.

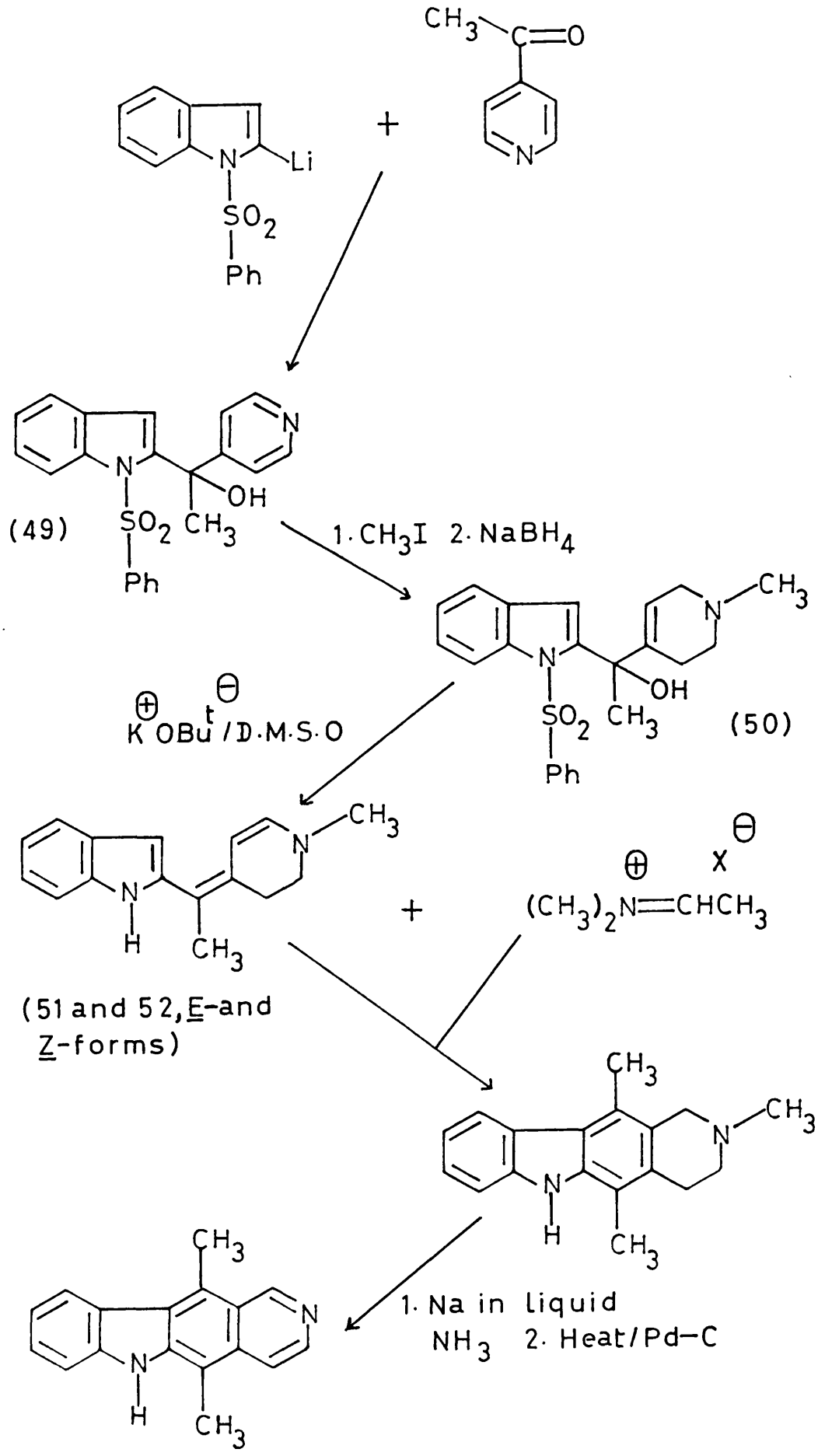
The alkaloid olivacine, which is as cytotoxic as ellipticine was first prepared by Schmutz and Wittwer[24], and a slightly modified route outlined in (Scheme 13), was used by Moscher et al[10] to prepare this alkaloid and certain of its derivatives for pharmacological evaluation. Since then a large amount of related work has been carried out[25,26,27,28].

Potier and his colleagues[29a,b] in France have devised three routes to ellipticine which are of special interest because they are considered by the authors to resemble, to a certain extent, the biosynthetic pathway by which the alkaloid is formed in nature. The first is shown in (Scheme 14).

Scheme 13.



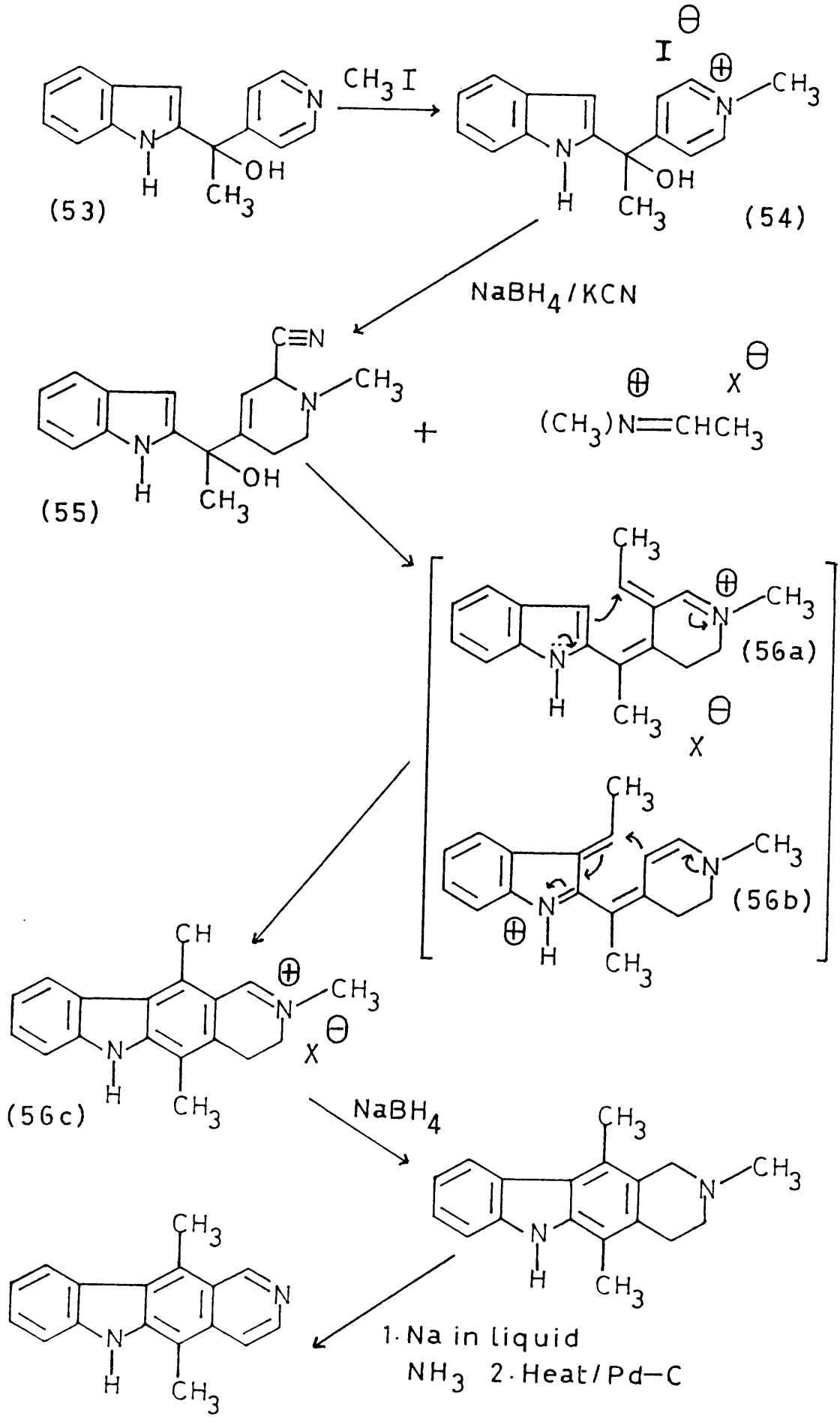
Scheme 14.


CH3
C=O
N
Li
SO2
Ph
+
N
OH
(49)
SO2
CH3
Ph
1. CH3I 2. NaBH4
CH3
N
OH
SO2
CH3
Ph
(50)
K⊕ OBut⊖ / D.M.S.O
CH3
N
N
H
CH3
(51 and 52, E-and Z-forms)
+
(CH3)2N=CHCH3
X⊖
CH3
CH3
N
N
H
CH3
1. Na in liquid NH3 2. Heat/Pd–C
CH3
N
N
H
CH3

In this approach the required 2-substituted indole (49) was prepared by the condensation of 2-lithio-1-sulphobenzoylindole with 4-acetylpyridine. Quaternisation was effected with iodomethane and the methiodide salt reduced with sodium borohydride, thus affording the tetrahydropyridine (50). When treated with potassium tertiary butoxide in dimethylsulphoxide this yielded the isomeric (E- and Z)-dienamines (51 and 52, only one of which is shown). Reaction with the Mannich reagent formed from methylamine and acetaldehyde gave 2-methyl-1,2,3,4-tetrahydroellipticine, which was demethylated using sodium in ammonia and dehydrogenated over palladium carbon at elevated temperature in an inert solvent. The yield from (50) was 2.2%.

The second method outlined in (Scheme 15), involved the hydrolysis of (49) to the parent indole (53) and reduction of the corresponding iodomethylate (54) with sodium borohydride in the presence of a large excess of potassium cyanide. The product (55) was treated with the same Mannich reagent as described in (Scheme 14)to give the imminium salt (56c), presumably by way of the intermediate (56a or 56b). This was reduced without isolation using sodium borohydride to give 2-methyl-1,2,3,4-tetrahydroellipticine. Finally demethylation and dehydrogenation steps were carried out as before in (Scheme 14). The yield from the cyanide (55) was 24%.

Scheme 15.

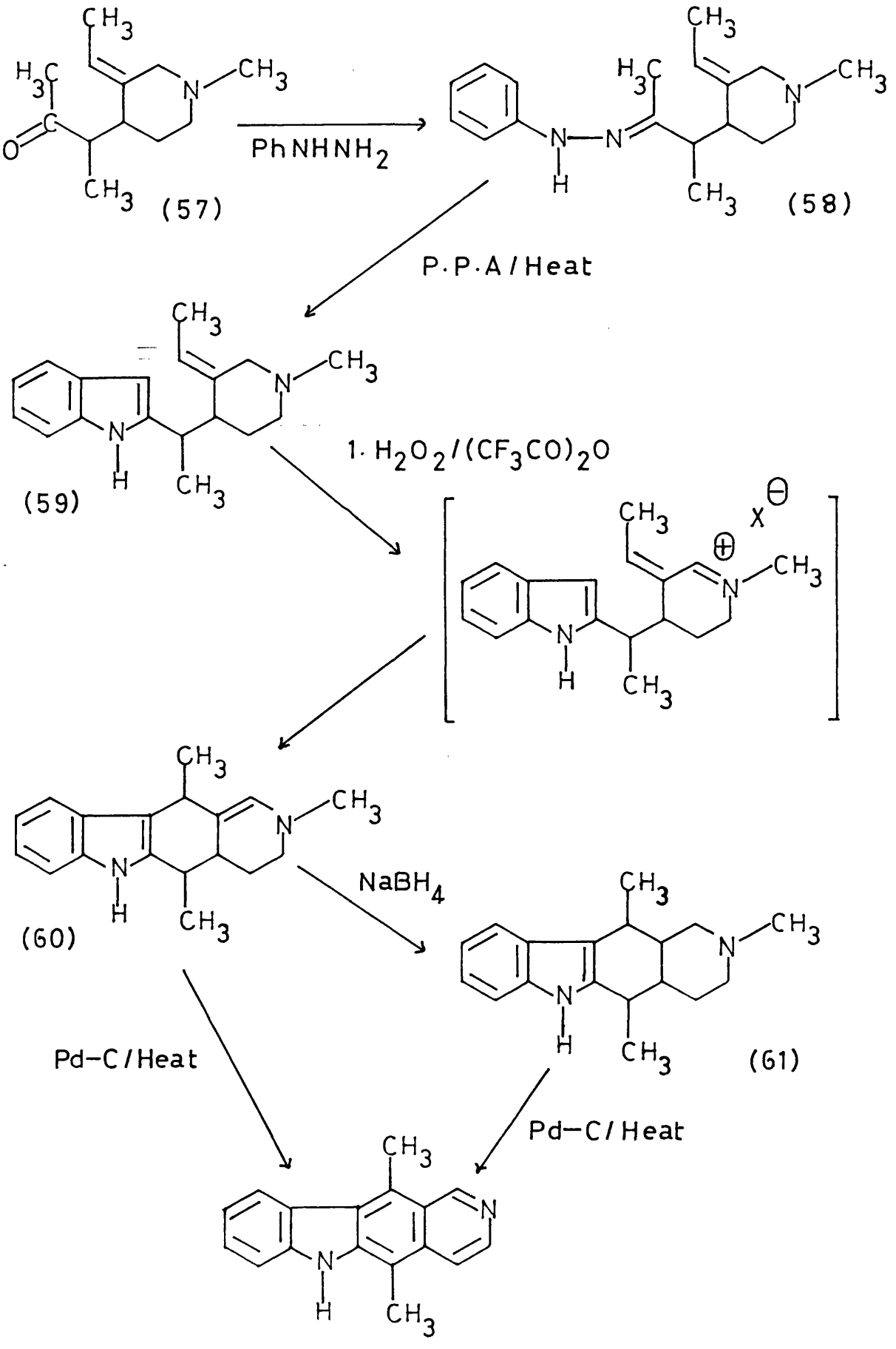

CH3I
(53)
(54)
NaBH4/KCN
(55)
(CH3)N=CHCH3
(56a)
(56b)
(56c)
NaBH4
1·Na in liquid
NH3 2·Heat/Pd–C

The third method 29[b] (Scheme 16), involved the formation of the phenylhydrazone (58) of the ketone (57), and its conversion *via* a Fischer reaction to the indole (59). This was cyclized with hydrogen peroxide and trifluoroacetic anhydride to give 2,3,4,4a,5,11-hexahydro-2-methylellipticine (60). The tetracyclic system was reduced to the octahydro compound (61) with sodium borohydride. Aromatization and demethylation was then achieved by heating with palladium carbon in an inert solvent. The aromatization of the hexahydropyridocarbazole (60) may also be effected directly. Both methods giving an overall yield, based on the phenylhydrazone of 18-20%.

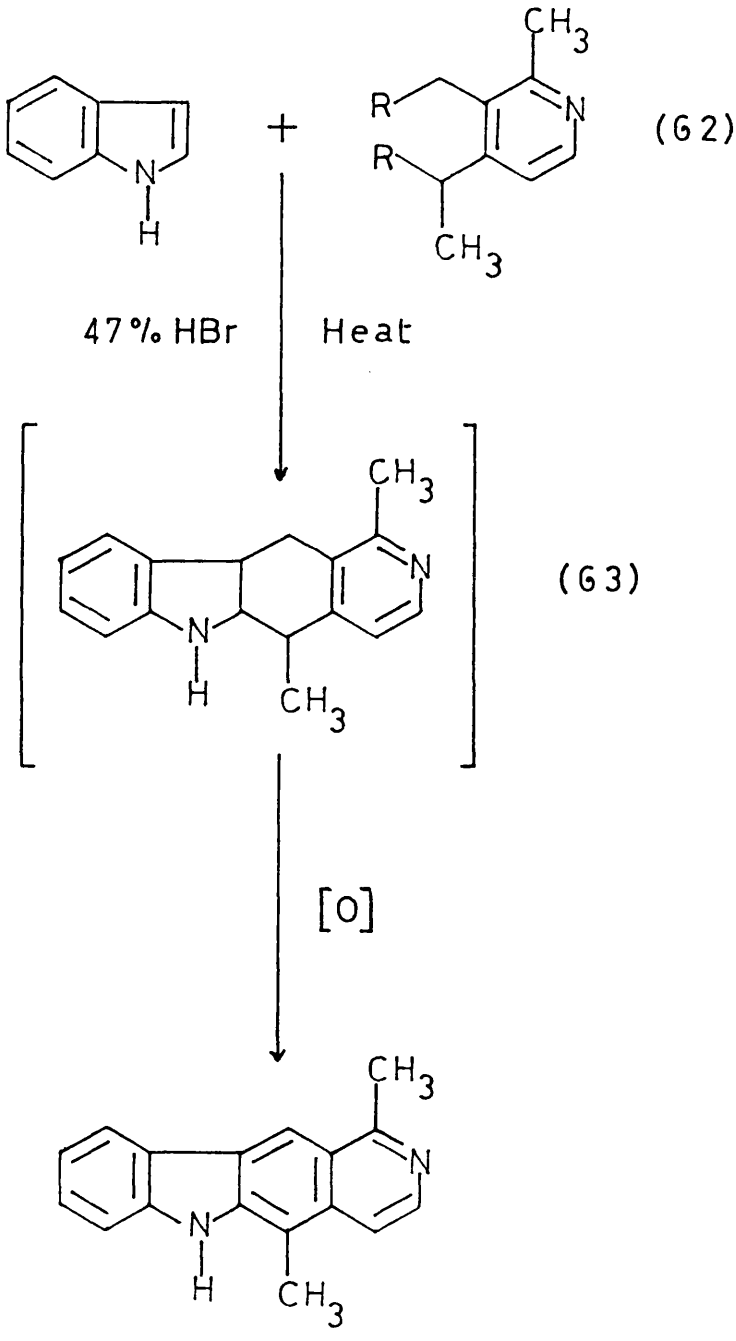
These routes although attractively short, suffer from the disadvantage of rather inaccessible starting materials, and once again, they have no utility when thermo-labile substituents are present.

Kametani and his team have also devised a number of interesting routes to certain 6*H*-pyrido[4,3-b]carbazoles, firstly they reported a so called 'one step' regiospecific synthesis of olivacine, (Scheme 17)[30a].

Scheme 16.


PhNHNH2
(57)
(58)
P.P.A / Heat
(59)
1. H2O2 / (CF3CO)2O
NaBH4
(60)
(61)
Pd–C / Heat
Pd–C / Heat

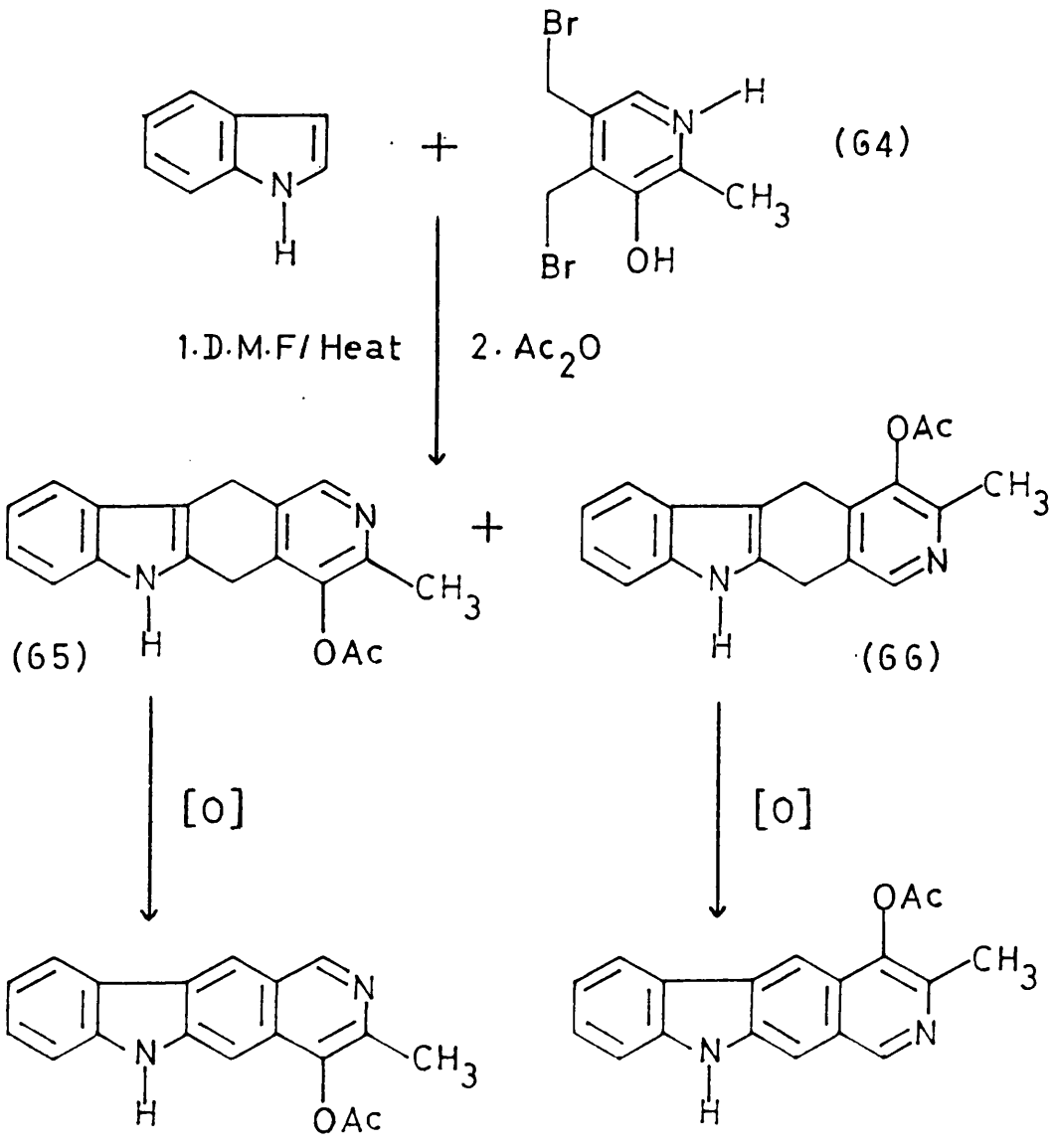
Scheme 17.



The claim of a one step synthesis is hard to justify because the 4-(1-hydroxyethyl)-3-hydroxymethyl-2-methylpyridine (62, R = OH), was itself prepared by a multistage synthesis. However, (62) was condensed with indole by heating in 47% aqueous hydrobromic acid. The corresponding dibromo compound (62, R = Br), was formed <u>in situ</u> and reacted to give olivacine, presumably <u>via</u> the dinydro intermediate (63). The yield was 30% overall.

The second approach[30b] is outlined in (Scheme 18).

<u>Scheme 18</u>

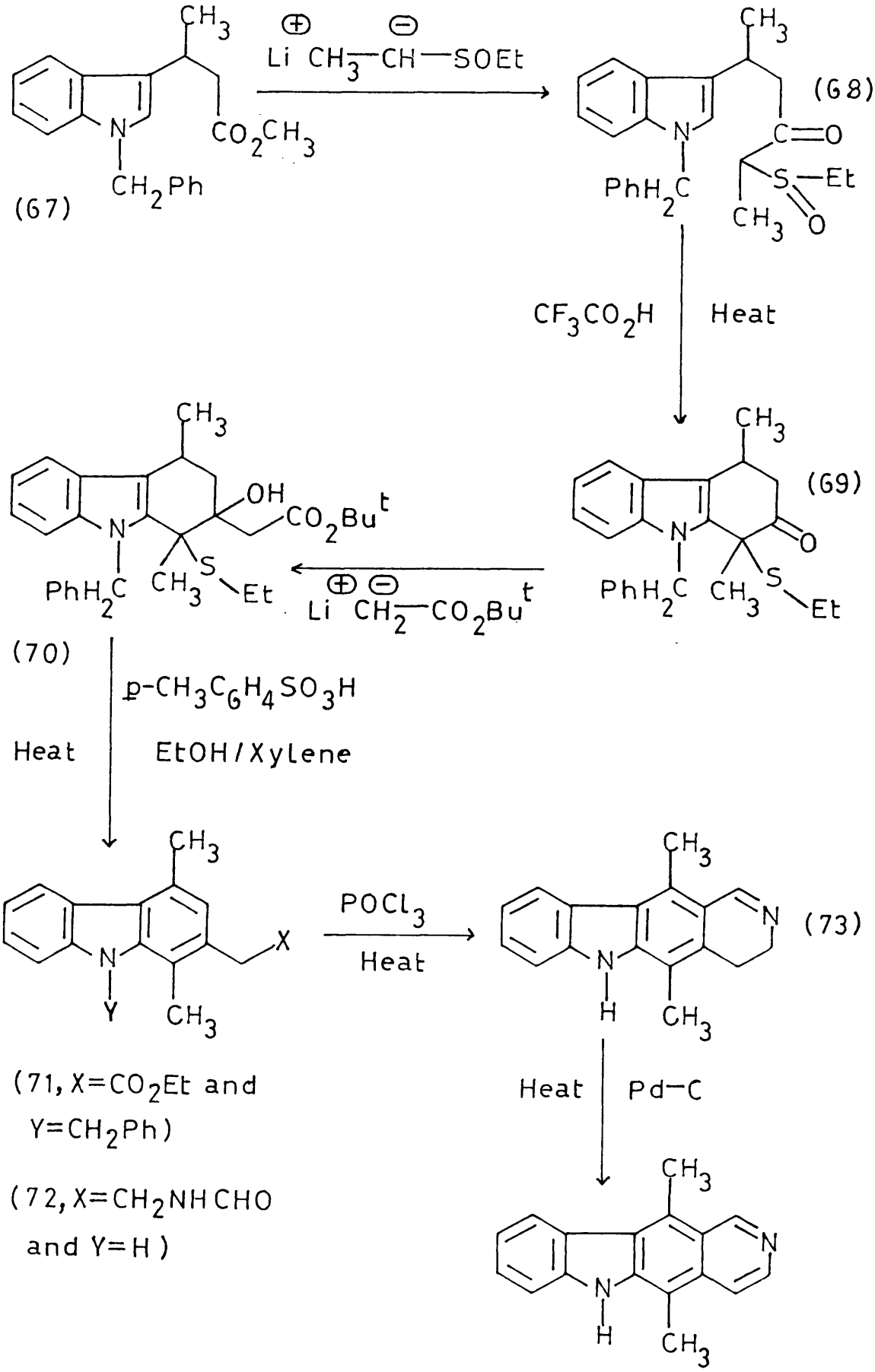


Here indole was condensed with 4,5-dibromoethyl-3-hydroxy-2-methylpyridinium bromide (64) by warming in dimethylformamide and acetylated using acetic anhydride to give a mixture of the 5,11-dihydropyrido[4,3-b] carbazole (65) and its structural isomer (66). These were left to stand in air and slowly oxidised to the fully aromatic compounds. The yields of the isomeric products were (65) 4% and (66) 15%. Once again this synthesis is bedevilled by inaccessible starting materials and low yields.

Another Japanese publication[28] describes the synthesis of ellipticine from methyl-1-benzylindole-3-butyrate (67) outlined in (Scheme 19).

Treatment of methyl-1-benzylindole-3-butyrate (67) with the lithium salt of diethylsulphoxide gave the compound (68), which cyclized on heating with trifluoroacetic acid to give (69). Attack on the carbonyl group of this compound by t-butyl lithioacetate gave (70) as a mixture of stereoisomers, from which the carbazole (71) was obtained by treatment with p-toluenesulphonic acid in a boiling xylene/ethanol mixture. A series of steps (c.f. Govindachari's method p. 9) converted this material into N-(1,4-dimethyl-9H-carbazol-2-yl)ethyl formamide (72). The compound (72) was ring closed using a modified Bischler-Napieralski reaction to give the 3,4-dihydro derivative (73), which was dehydrogenated by heating with palladium on charcoal. The overall yield of ellipticine was 23%, but this route has the disadvantage of including too many steps for it to be considered as a practical method for the synthesis of substituted derivatives.

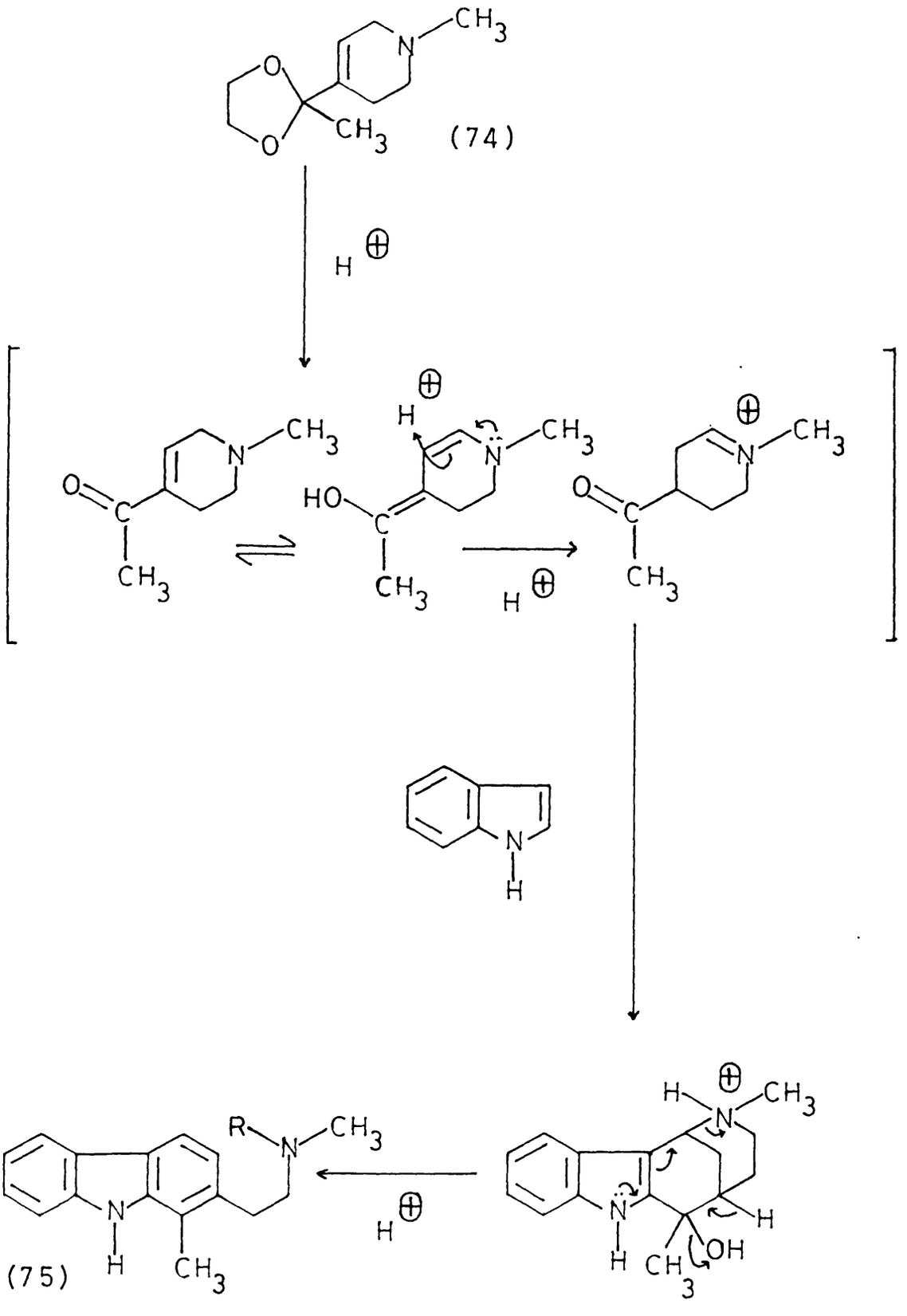
Scheme 19.

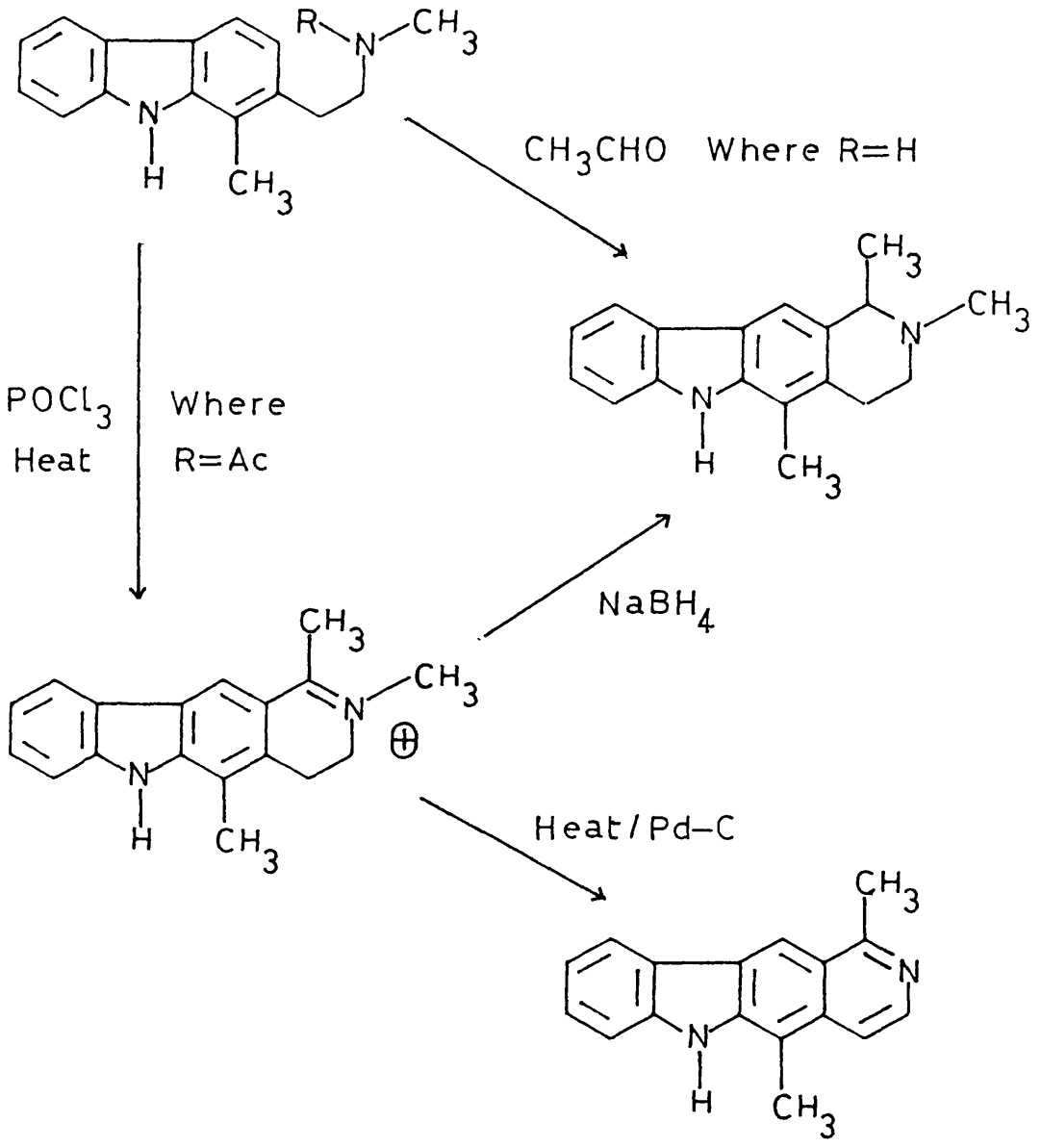

(67)
(68)
(69)
(70)
(71, X=CO2Et and Y=CH2Ph)
(72, X=CH2NHCHO and Y=H)
(73)
Heat
CF3CO2H
Heat
p-CH3C6H4SO3H
EtOH/Xylene
POCl3
Heat
Heat
Pd-C

An attractive synthesis of (±)-guatambuine ($N_{(b)}$-methyl-1,2,3,4-tetrahydro olivacine) has been published[31], and is outlined in (Scheme 20). Indole was condensed with the ketal (74) in boiling 50% aqueous acetic acid to give the 1,2-disubstituted carbazole (75). This presumably forms *via* the mechanism shown. The carbazole (75) was reacted with acetaldehyde to afford (±)-guatambuine directly in 26% yield from indole.

Alternatively the acetyl derivative (75, R = Ac) was subjected to a Bischler-Naperialski reaction using phosphorus oxychloride to give an iminium salt. Reduction of this with sodium borohydride gave (±)-guatambuine in 45% yield, which is more efficient than the direct method. The iminium salt was also dehydrogenated and demethylated directly to olivacine by heating over palladium carbon in an inert solvent.

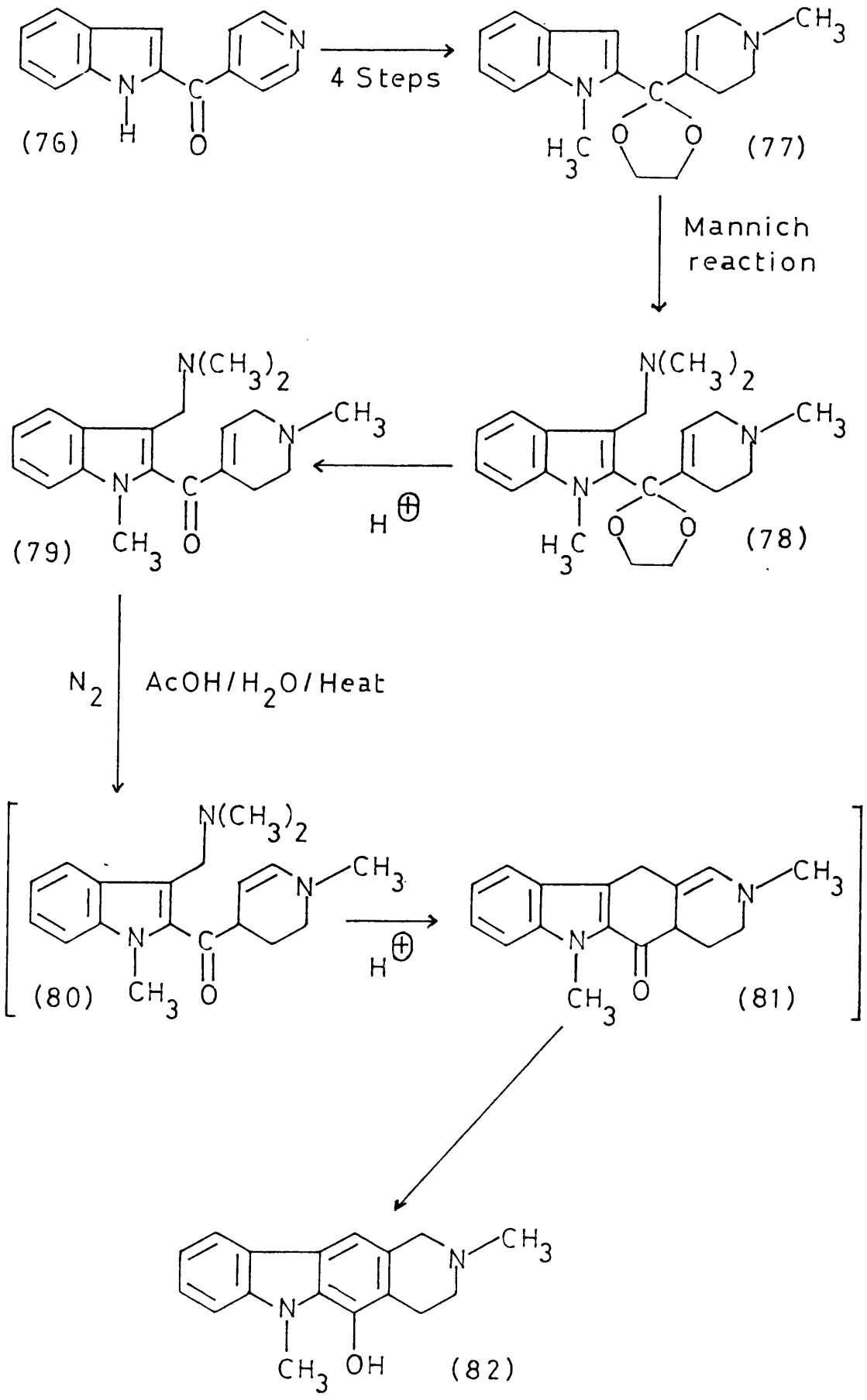
Scheme 20.





Due to the very encouraging anti-cancer activity of 9-hydroxyellipticine compared with unsubstituted ellipticine, considerable synthetic effort has been expended to prepare other hydroxylated derivatives for pharmacological testing. Martinez and Joule[32] have synthesized the 5-hydroxypyridocarbazole (82) by the method outlined in (Scheme 21).

Scheme 21.


(76)
4 Steps
(77)
Mannich reaction
(78)
H⊕
(79)
N2
AcOH/H2O/Heat
(80)
H⊕
(81)
(82)

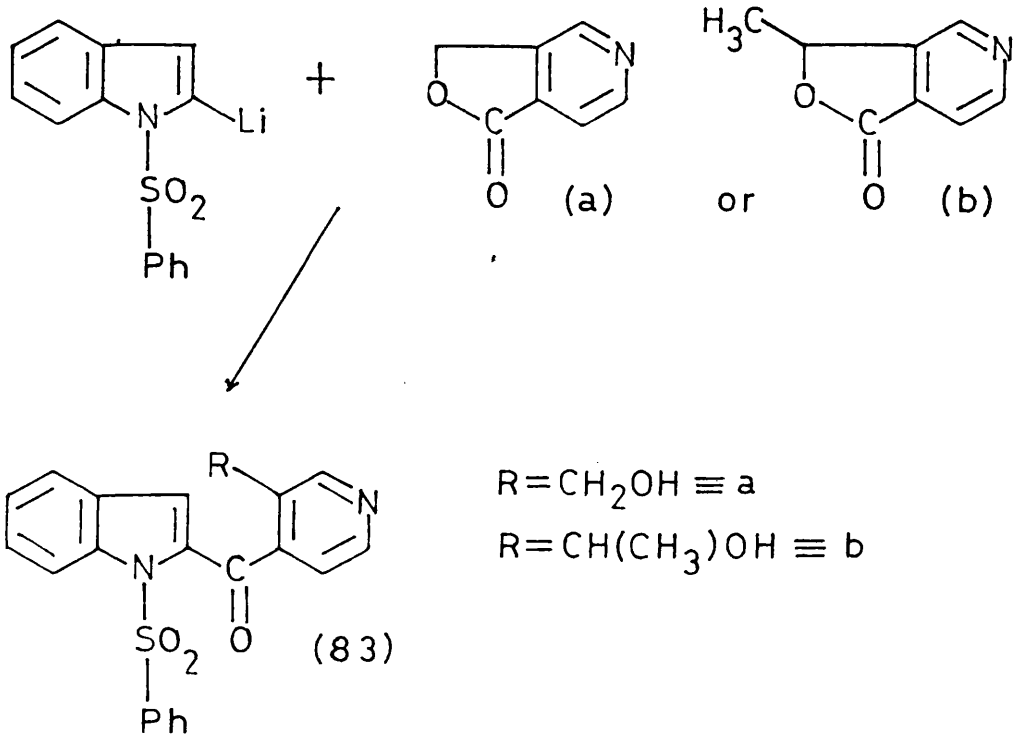
The compound (77) was prepared from the known indole (76) by protecting the carbonyl group through acetalisation with ethylene glycol, followed by N-methylation of the pyridine and indole moieties. Partial reduction of the pyridine ring was then achieved with sodium borohydride in boiling ethanol. The overall yield for the four steps was 70%.

A Mannich reaction was carried out on (77) to give the 3-substituted indole (78) and the ketal group was then removed with 5N-hydrochloric acid to yield the ketone (79). This was heated in 5% aqueous acetic acid solution under a protective nitrogen atmosphere to give the hydroxylated tetracycle (82), in 25% yield. The reaction presumably occurs through the intermediates (80) and (81).

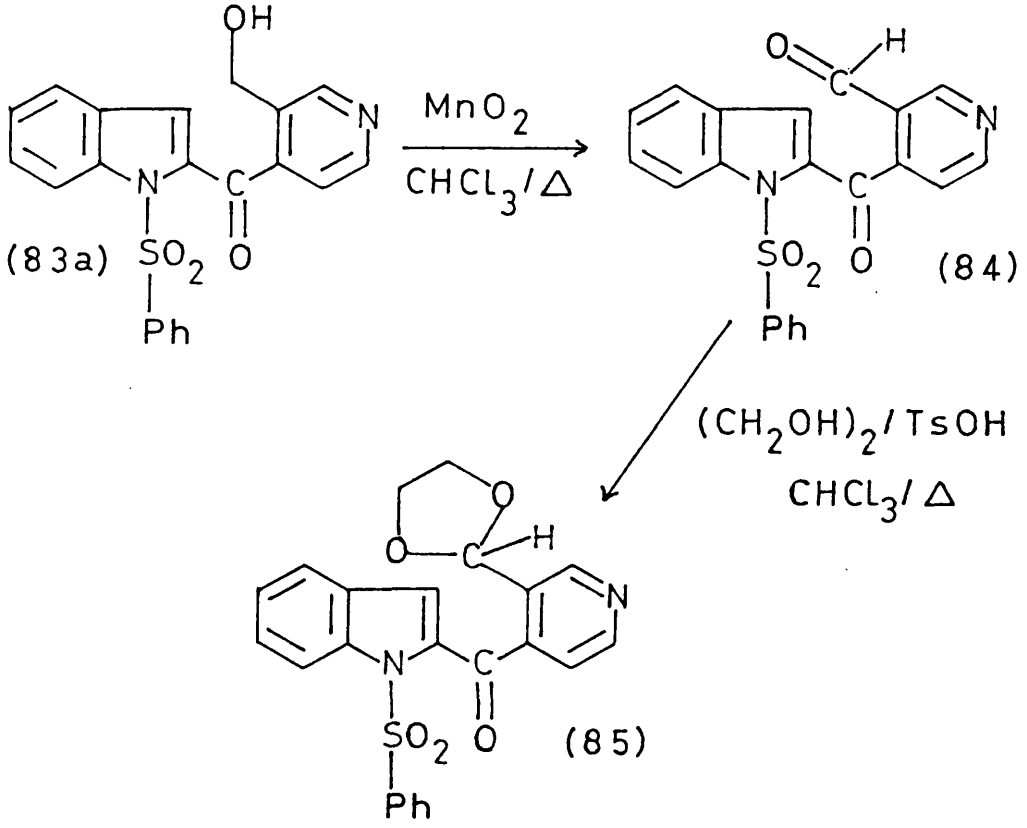
The final product still requires $N_{(b)}$-demethylation and dehydrogenation before the fully aromatic structure is formed. While this could be accomplished it seems unlikely that the yield would be sufficiently high to justify the use of this method as a general synthetic route.

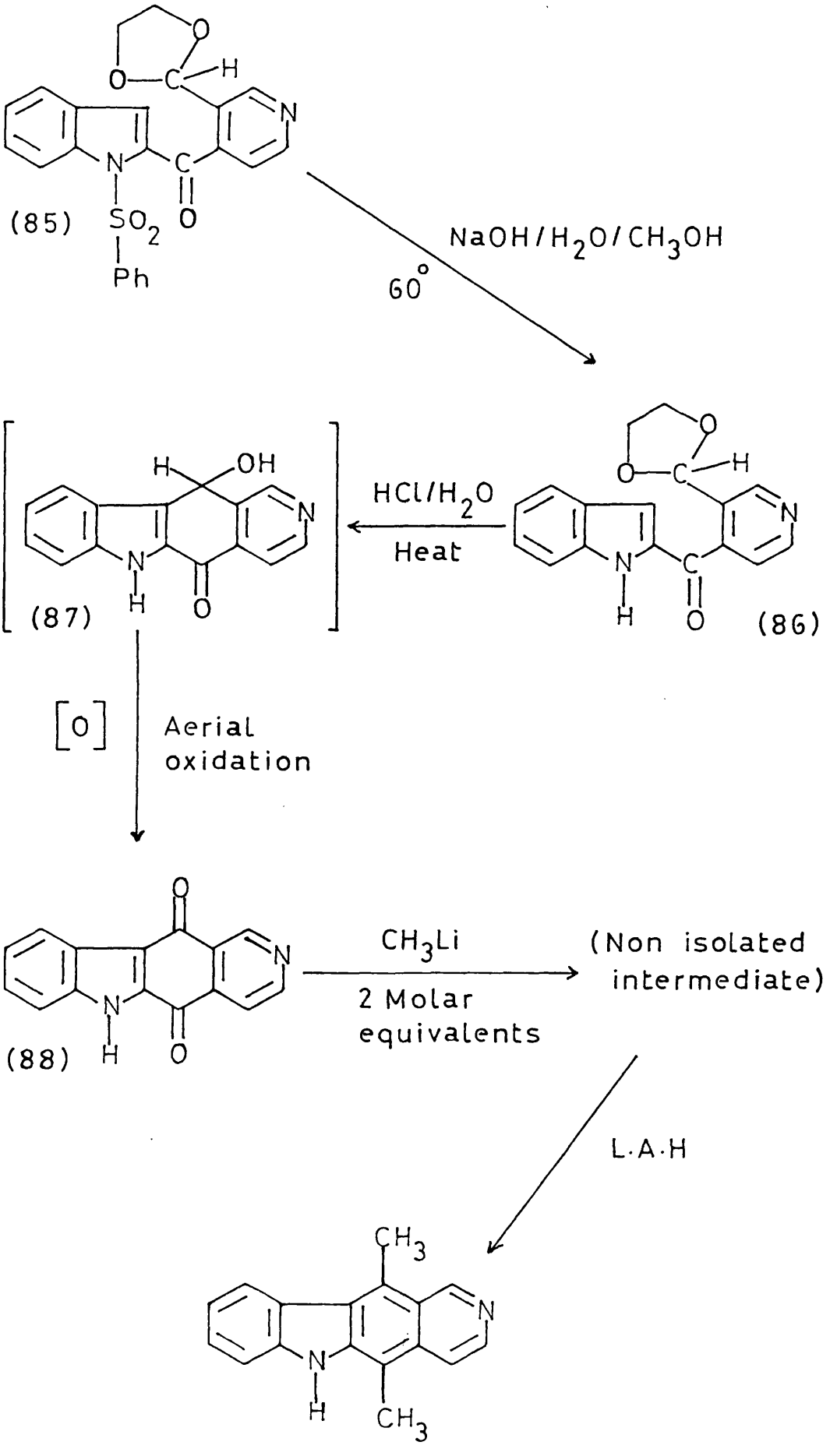
Further work by J.A. Joule and D.A. Taylor gives rise to two related syntheses of ellipticine.[33] These methods are of particular interest as both are reasonably short and have the potential for the preparation of side-chain modified analogues. These two routes are outlined in (Scheme 22).

Scheme 22.

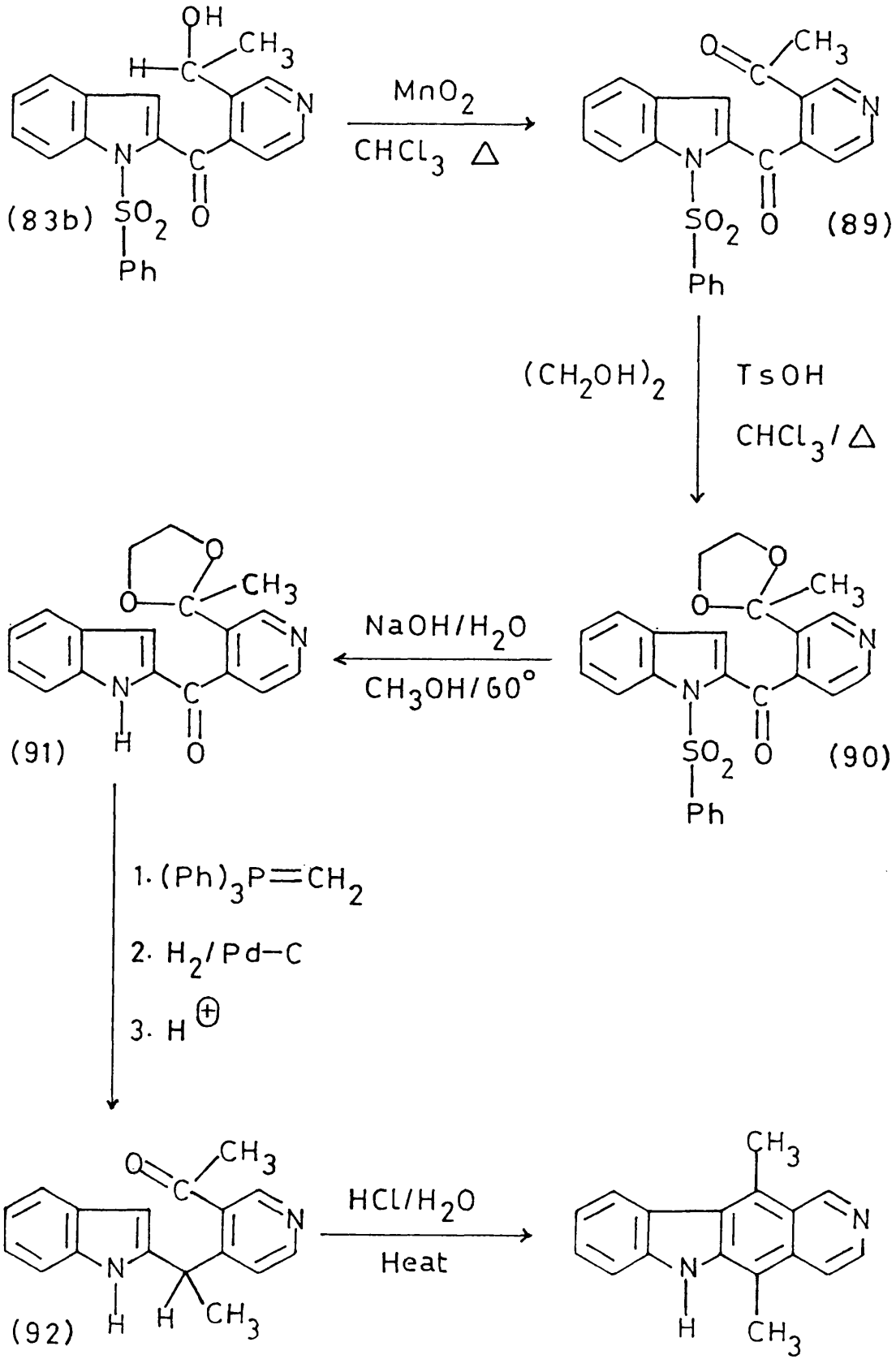


Route a




(85)
NaOH/H2O/CH3OH
60°
HCl/H2O
Heat
(87)
(86)
[O]
Aerial oxidation
(88)
CH3Li
2 Molar equivalents
(Non isolated intermediate)
L.A.H

Route b


(83b)
MnO2
CHCl3 Δ
(89)
(CH2OH)2
TsOH
CHCl3/Δ
(90)
NaOH/H2O
CH3OH/60°
(91)
1. (Ph)3P=CH2
2. H2/Pd–C
3. H⊕
(92)
HCl/H2O
Heat

Route a

Condensation of 2-lithio-1-benzenesulphonylindole with the pyridine lactones (a) and (b) gave the keto alcohols (83a) and (83b) respectively.

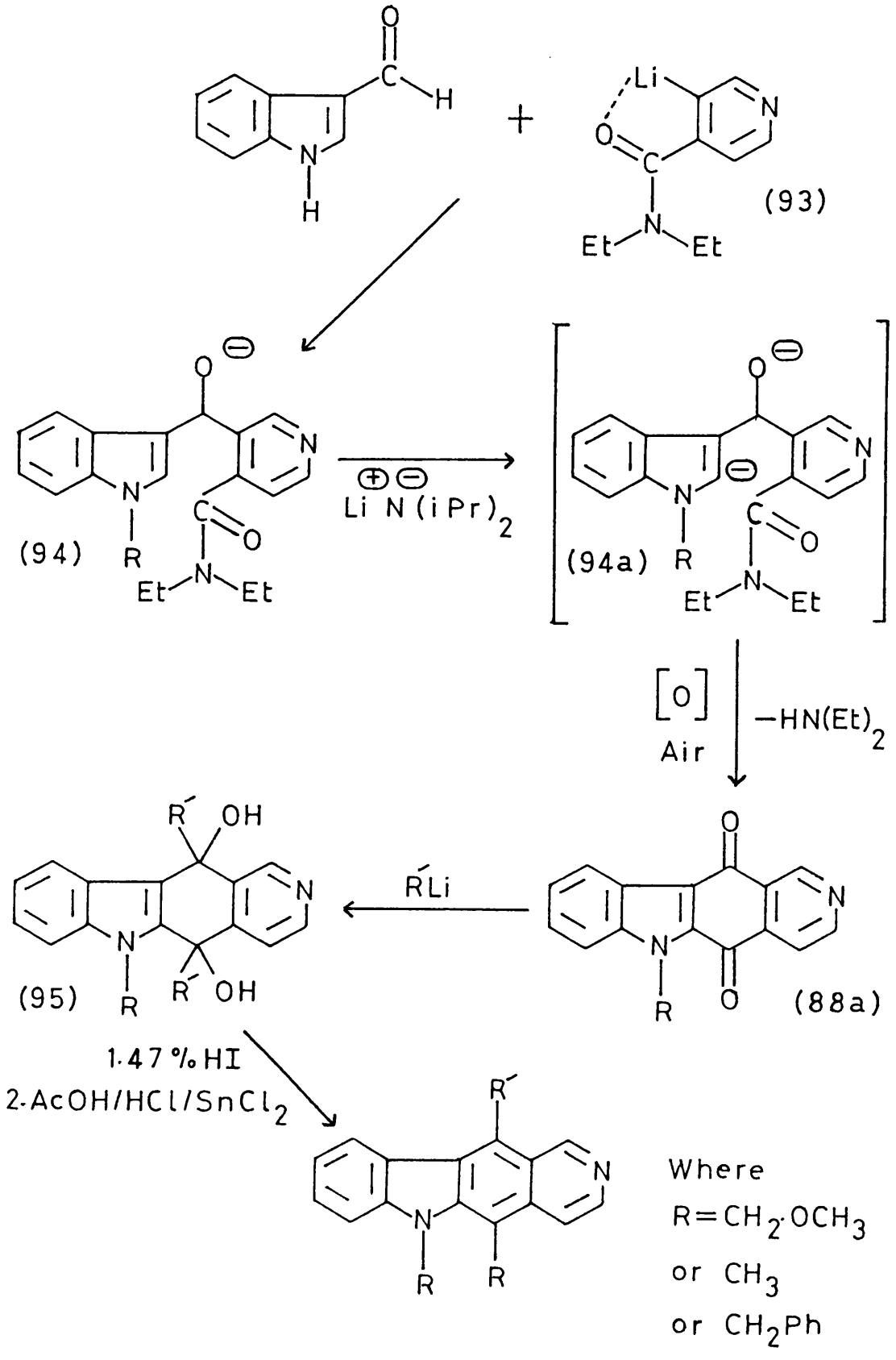
The primary alcoholic group of (83a) was oxidised with manganese dioxide to give the keto-aldehyde (84). This was selectively acetalised under mild conditions to give the acetal (85). Alkaline removal of the $N_{(a)}$-protecting group gave the keto acetal (86). This was followed by hydrochloric acid catalysed removal of the acetal which also brought about condensation at the indole β-position. The reaction presumably occurs through the intermediate (87), but this was not isolated because aerial oxidation gave the dark red quinone (88). This quinone was treated with three molar equivalents of methyl lithium and the product reduced immediately without isolation using lithium aluminium hydride to give ellipticine.

Route b.

The secondary alcohol (83b) was oxidised with manganese dioxide to the ketone (89) and this selectively acetalised to yield the acetal (90). N-Deprotection of the acetal was achieved with sodium hydroxide, affording the keto acetal (91). A Wittig reaction was then carried out using triphenylphosphonium methylide, followed by catalytic reduction and treatment with acid to give the pyridyl ketone (92). This compound has the appropriate substitution and oxidation level for conversion to ellipticine, which was achieved by the action of dilute hydrochloric acid at reflux temperature.

Another successful approach to the ellipticine skeleton that also employed the quinone (88), was that of Snieckus[34], and his group in Canada, whose route is outlined in (Scheme 23).

Scheme 23.

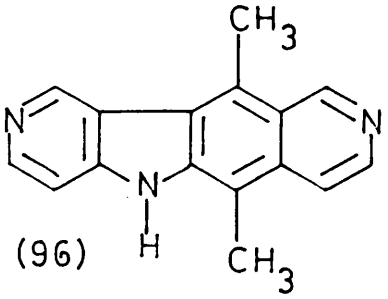


N-Methylindole-3-carboxaldehyde was condensed with 3-lithiopyridine-4-diethylamide (93) to give the amide (94). Treatment of this with lithium diisopropylamide removed a proton from the indole-2-position to give the dianion (94a). This dianion cyclized spontaneously to give a product that was not isolated, but directly exposed to aerial oxidation, which gave rise to the same type of quinone system (88a) that has been independently synthesized by Joule[33]. Treatment of the quinones (88a) with methyl lithium gave the intermediates (95, $R^1 = CH_3$), which were subsequently reduced to the required ellipticine analogues under acid conditions. In the case of the methylenemethoxy derivative (95, $R^1 = CH_3$, $R = CH_2OCH_3$), the powerful reductive acidic conditions used, gave ellipticine directly. With a wide range of alkyl lithium reagents available this route is entirely flexible in allowing the synthesis of other 5,11-substituted pyrido[4,3-b]carbazoles and their analogues.

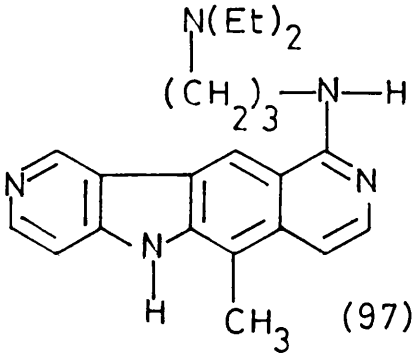
Ellipticine and its analogues are considered to exert their anti-tumour activity by intercalation between the D.N.A. base pairs of cancer cells, (see pharmacological section p.222). A relationship exists between the pKa of the intercalating molecule and its affinity constant for D.N.A. The higher the pKa value, the more likely is the structure to intercalate across D N.A. base pairs.

The synthesis of new structural types (96) and (97) in which the A ring of ellipticine is replaced by a pyridine

nucleus have been described by Bisagni and his associates[35]. It was hoped that the extra nitrogen atom in the system, which confers a higher pKa value, would make these compounds more effective against cancer cells.



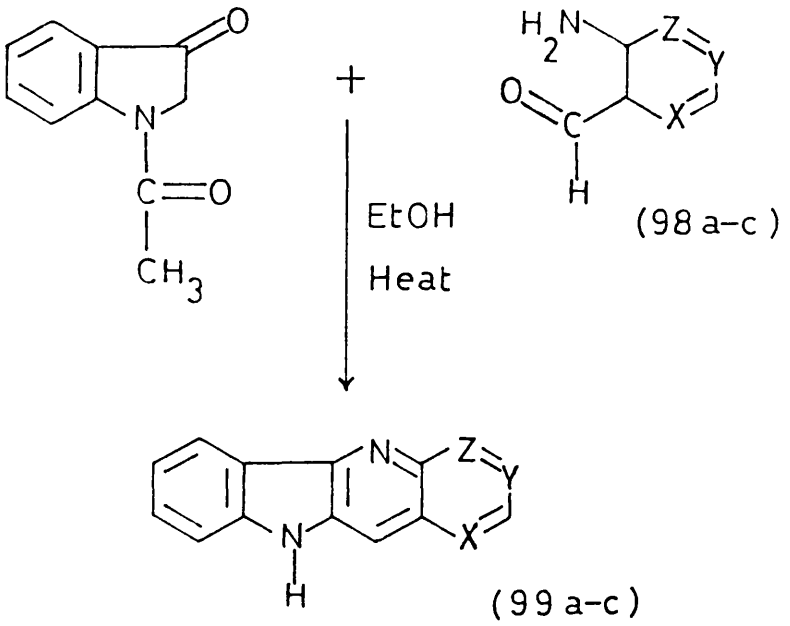
(96)

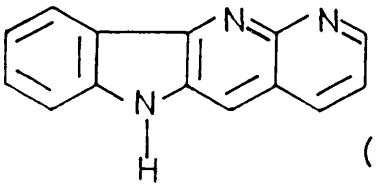


(97)

However, on pharmacological evaluation these compounds were found to have closely similar biological properties to ellipticine but do not appear to offer any therapeutic advantages.

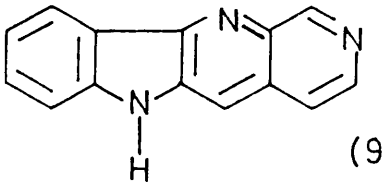
Similarly Queguiner et al[36], in France, have extended this approach by the synthesis of the 6H-indolo[3,2-b]napthyridines (99a-c). This was achieved by condensation of the aminoformyl pyridines (98a-c) with N-acetyl indoxyl, using an extension of the Friedlander quinoline synthesis, first applied to napthyridines by Hawes[39].





(99a)

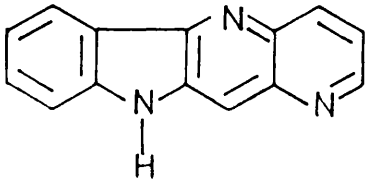
Where Z=N and X=Y=CH



(99b)

Where Y=N and Z=X=CH

(99c)

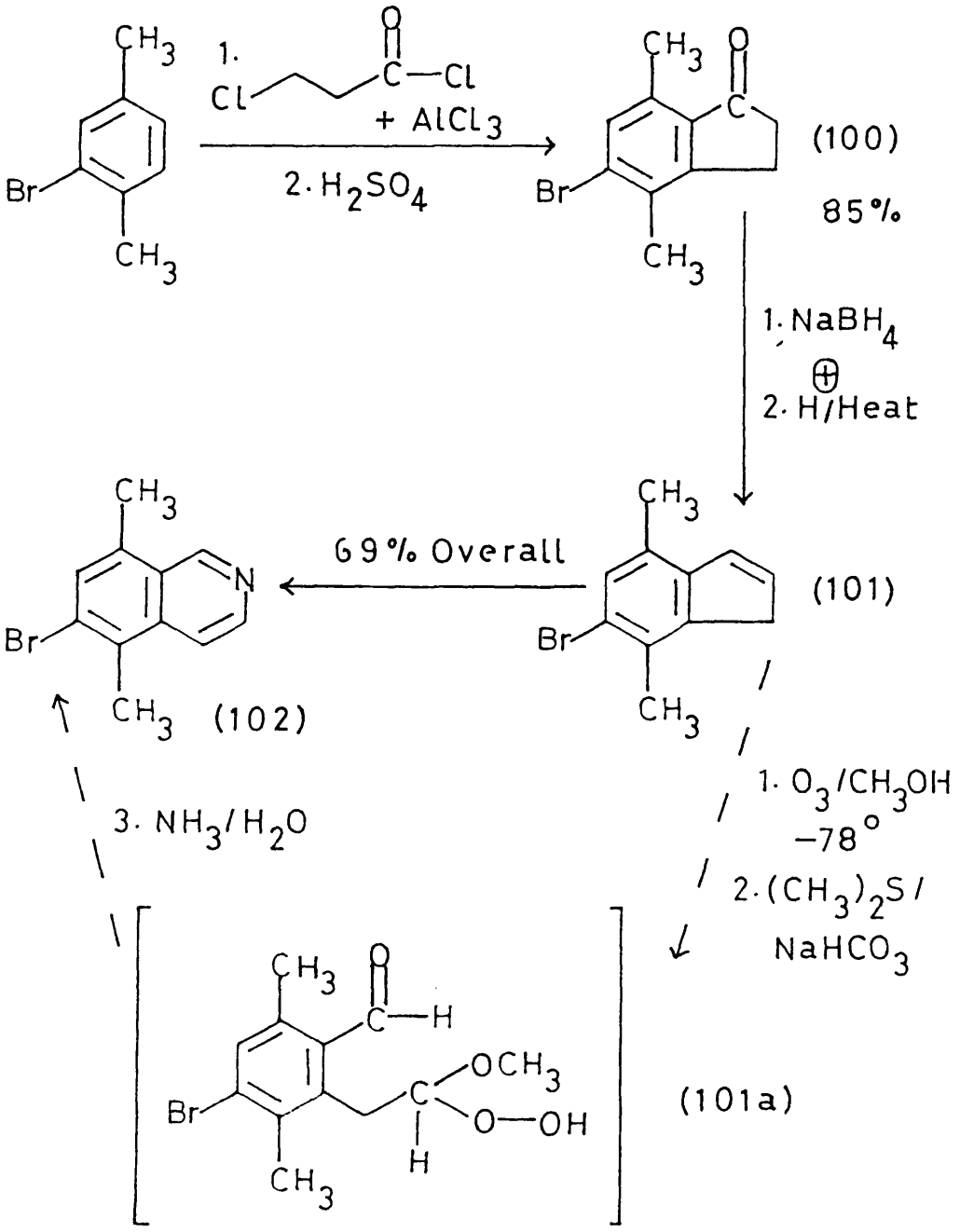


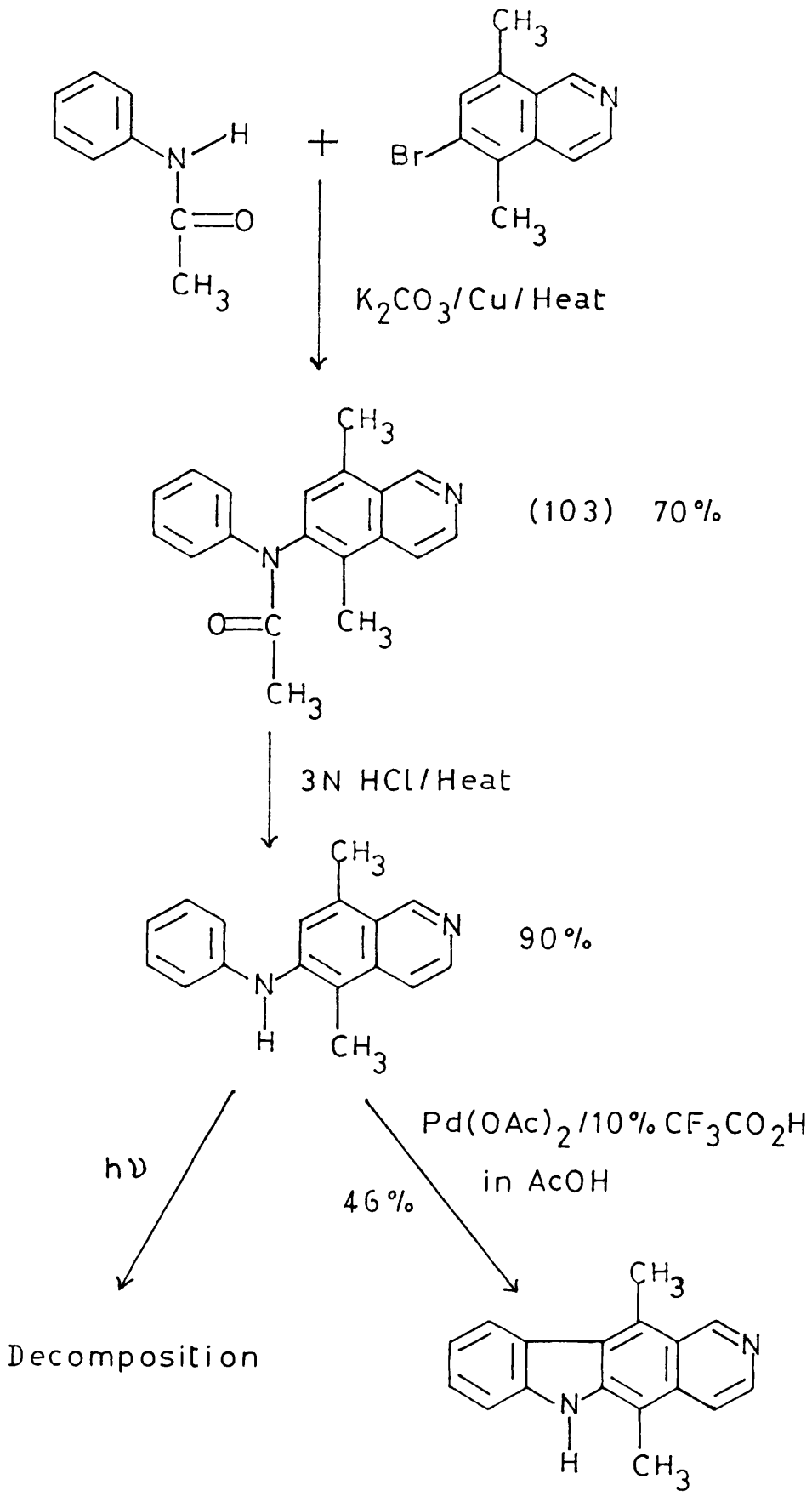
Where X=N and Z=Y=CH

The condensation was simply achieved by heating the reactants together in ethanol at reflux temperature and the aminoformyl pyridines were prepared from the corresponding aminopyridine carboxylic acids by the methods of Armarego[37] and Hawes[38].

Miller and Mook[40] have recently derived an interesting new approach to ellipticine. These workers chose to synthesize the isoquinoline (rings 'C' and 'D') portion of the molecule first. They prepared the bromoisoquinoline (102) and coupled this with acetanilide. The heterocyclic ring was then closed using palladium acetate to give ellipticine as outlined in (Scheme 24).

Scheme 24.




CH3
N
H
Br
C=O
CH3
CH3
K2CO3/Cu/Heat
(103) 70%
3N HCl/Heat
90%
hν
Pd(OAc)2/10% CF3CO2H
in AcOH
46%
Decomposition
15—25% Overall

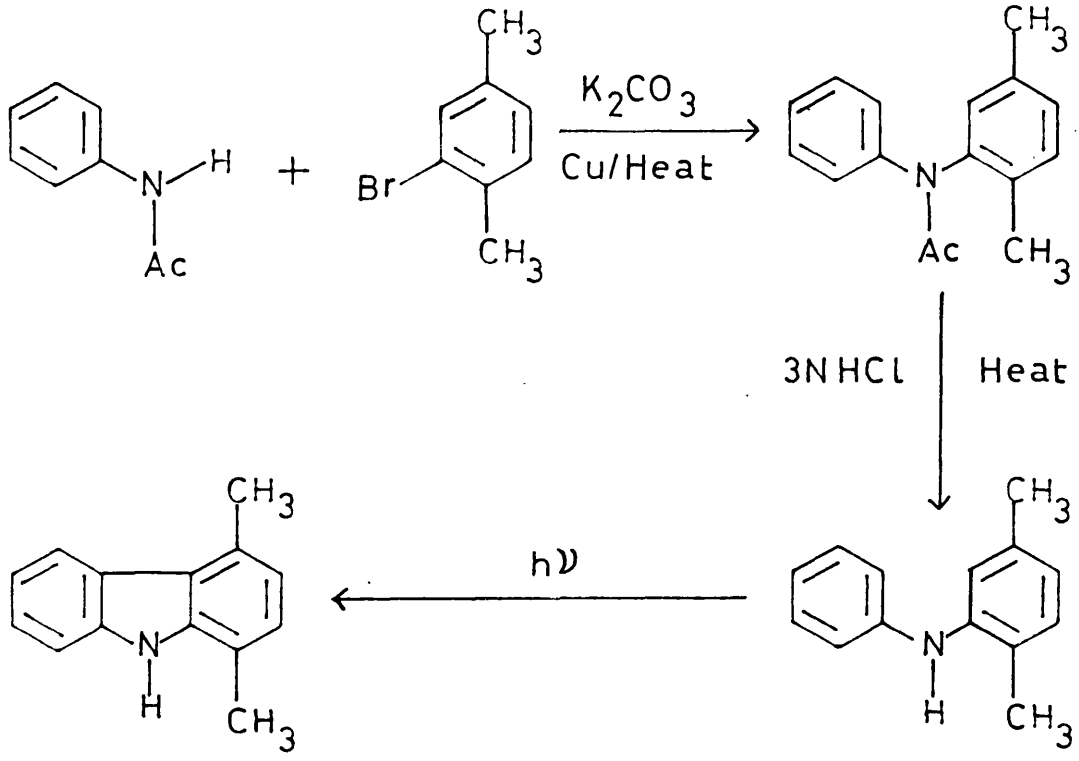
Synthesis of 6-bromo-5,8-dimethylisoquinoline (102) was achieved by condensation of 2-bromo-p-xylene with 3-chloropropionyl chloride in the presence of aluminium chloride, followed by acid catalysed ring-closure to give 5-bromo-4,7-dimethylindan-1-one (100) in excellent yield. The indanone (100) was reduced with sodium borohydride, and the resulting alcohol subjected to acid catalysed dehydration to give the indene (101). Ozonolysis of this, followed by treatment with concentrated aqueous ammonia solution gave the fully aromatic isoquinoline (102). The reaction presumably proceeds through the intermediate aldehyde (101a)[41].

The next reaction was the coupling of the bromoisoquinoline (102) with acetanilide, using the Goldberg modification of the Ullmann coupling reaction,[42a] to give the diarylamide (103), which was hydrolysed under acid conditions to the desired 6-anilino-5,8-dimethylisoquinoline.

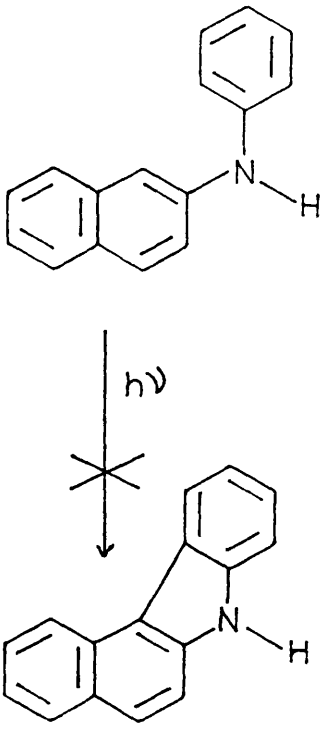
This compound was cyclized to the fully aromatic tetracycle using palladium acetate[43] in a trifluoroacetic acid/acetic acid solvent system.

The yield of this final step is 46% and the overall yield was 15-25%. This is comparable with many other ellipticine synthesis and the method should allow the preparation of some derivatives, although it does not offer strikingly significant advantages with respect to those discussed previously.

Other interesting facts were observed when photochemical cyclization was investigated. It is known that diarylamines give carbazoles upon photolysis[44] so the authors prepared 2-anilino-p-xylene as before and photolyzed it to 1,4-dimethylcarbazole in excellent yield.



This proved that the 1,4-dimethyl substitution did not represent a problem. However, when 6-anilino-5,8-dimethylisoquinoline was photolysed under similar conditions, no ellipticine was observed. It is also significant that N-phenyl-2-napthylamine does not give benzocarbazole on photolysis, see reference 42c.

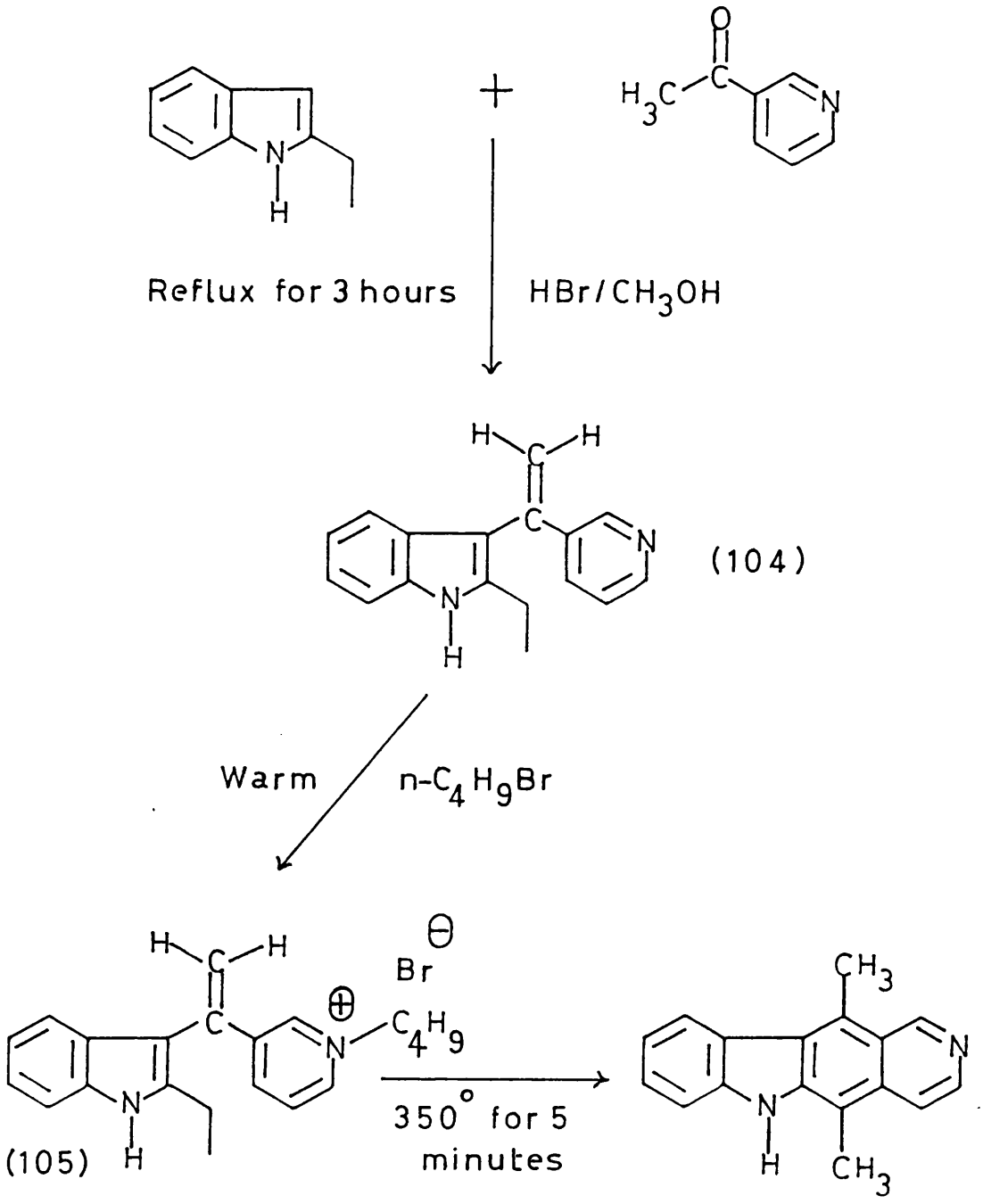


Miller and Mook[40], also claim that ellipticine proved to be photo-labile under the conditions used in their work, so they discontinued this approach.

Bergman and Carrlsson in Sweden[45] have found that it is possible to effect a 1:1 condensation between 2-ethylindole and 3-acetylpyridine. The 1:1 product is thought to arise because the bulky ethyl group sterically inhibits the formation of the bis-dindolyl compounds, which are always observed when indoles unsubstituted at the two position are used.

They have utilised this fact in a new and appealingly short ellipticine synthesis outlined in (Scheme 25).

Scheme 25.



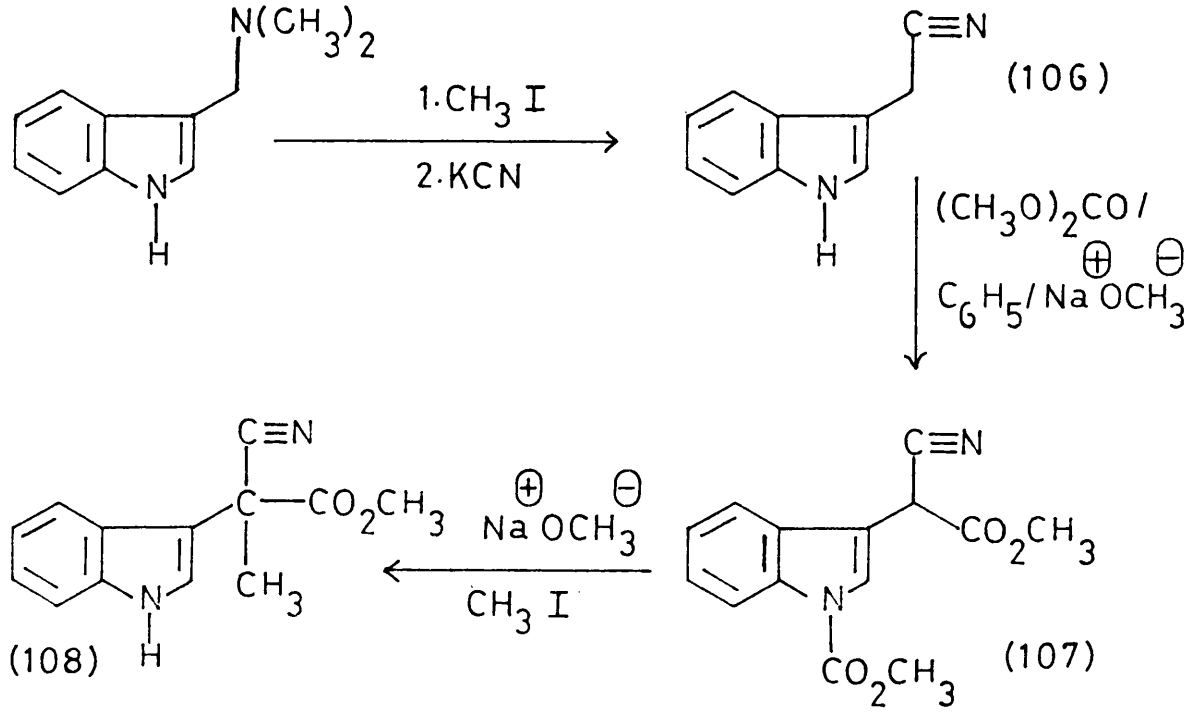
2-Ethylindole was prepared by condensing o-toluidine with propionic annhydride to give the appropriate amide, followed by the Madelung reaction to effect ring-closure with potassium tertiary butoxide at 185°.

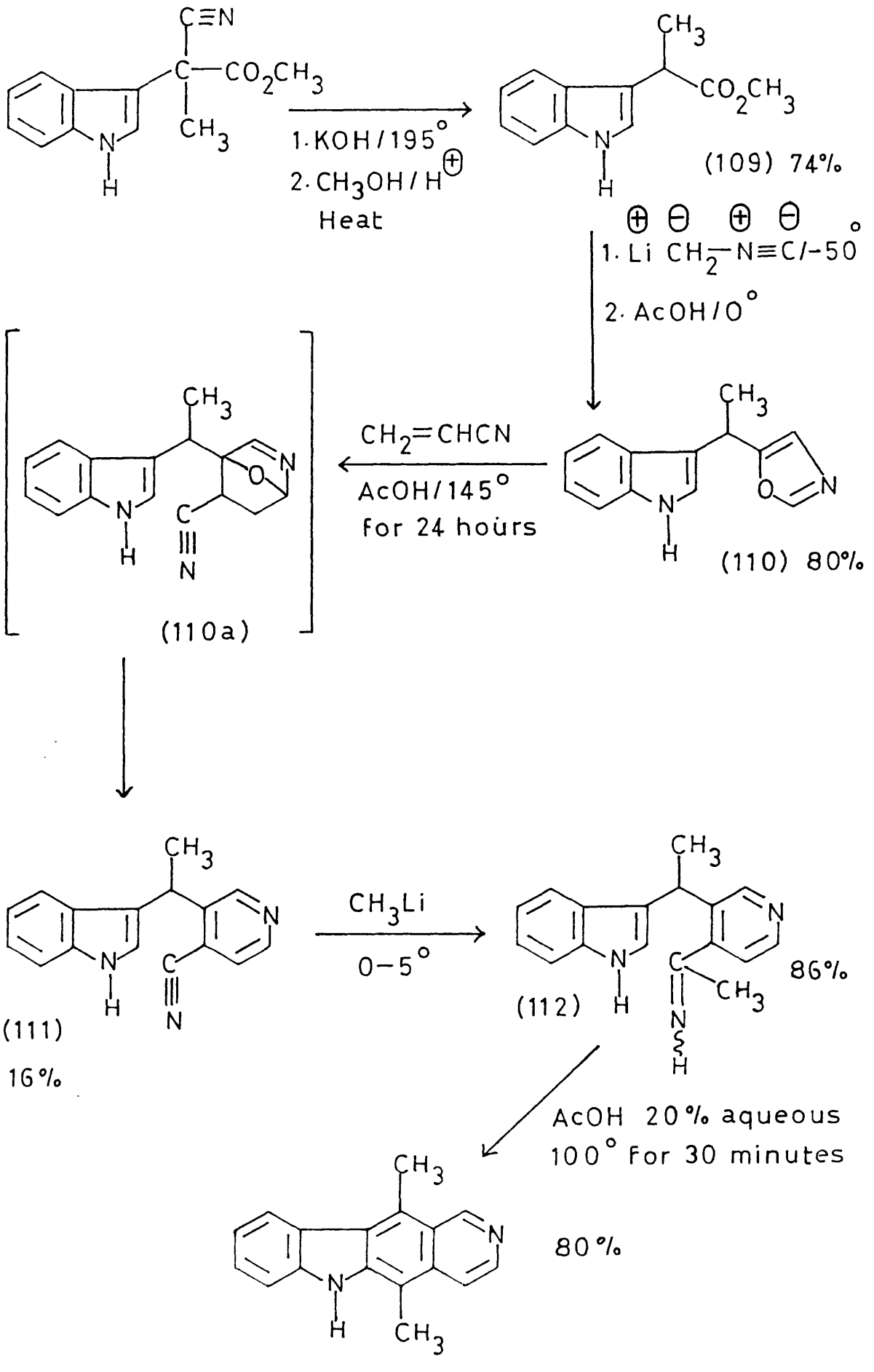
Equimolecular quantities of 2-ethylindole and 3-acetylpyridine were condensed by heating at reflux temperature in methanol saturated with hydrogen bromide to give 1-(3-indolyl-2-ethyl)-1-(3-pyridyl-ethylene (104). This compound was warmed with butyl bromide to give the quaternary salt (105), and pyrolysis of this at 350° for five minutes gave ellipticine.

While this synthesis is attractive from the point of view of readily obtainable starting materials and brevity, the ring closure conditions are extremely harsh, and this precludes its use for all but the most stable substituents.

Kozikowski and Hasan[46] have developed a strategy for ellipticine synthesis based on the formation of the pyridine ring *via* the Diels-Alder condensation of the azadiene portion of an oxazole with a dienophile[47]. This route is outlined in (Scheme 26).

Scheme 26.




1. KOH / 195°
2. CH3OH / H⊕
Heat
(109) 74%
1. Li CH2—N≡C / -50°
2. AcOH / 0°
CH2=CHCN
AcOH / 145°
for 24 hours
(110) 80%
(110a)
CH3Li
0–5°
86%
(111)
16%
(112)
AcOH 20% aqueous
100° for 30 minutes
80%

Indoleacetonitrile (106) was prepared from gramine by the action of methyl iodide, to give the quaternary base, followed by nucleophilic attack of cyanide ion[48]. The nitrile (106) was dicarbomethoxylated using dimethyl carbonate, sodium methoxide and benzene[49], to give (107). Further treatment with sodium methoxide and methyl iodide proceeded with loss of the N-carbomethoxy group and C-methylation to give the product (108). Hydrolysis and concomitant decarboxylation of (108) using potassium hydroxide in ethylene glycol at 195° followed by acid catalysed esterification with methanol[50], furnished the ester (109). Reaction of this ester with excess α -lithiated methyl isocyanide at -50°, followed by treatment with acetic acid at 0°[51] provided the oxazole (110), which was purified by chromatography on silica gel.

A Diels-Alder reaction of (110) with excess acrylonitrile in acetic acid at 145°, gave 3-[1-(indol-3-yl)ethyl]pyridine-4-carbonitrile (111) in 16% yield after two successive chromatographic purifications. The reaction presumably proceeds through the intermediate (110a)[52].

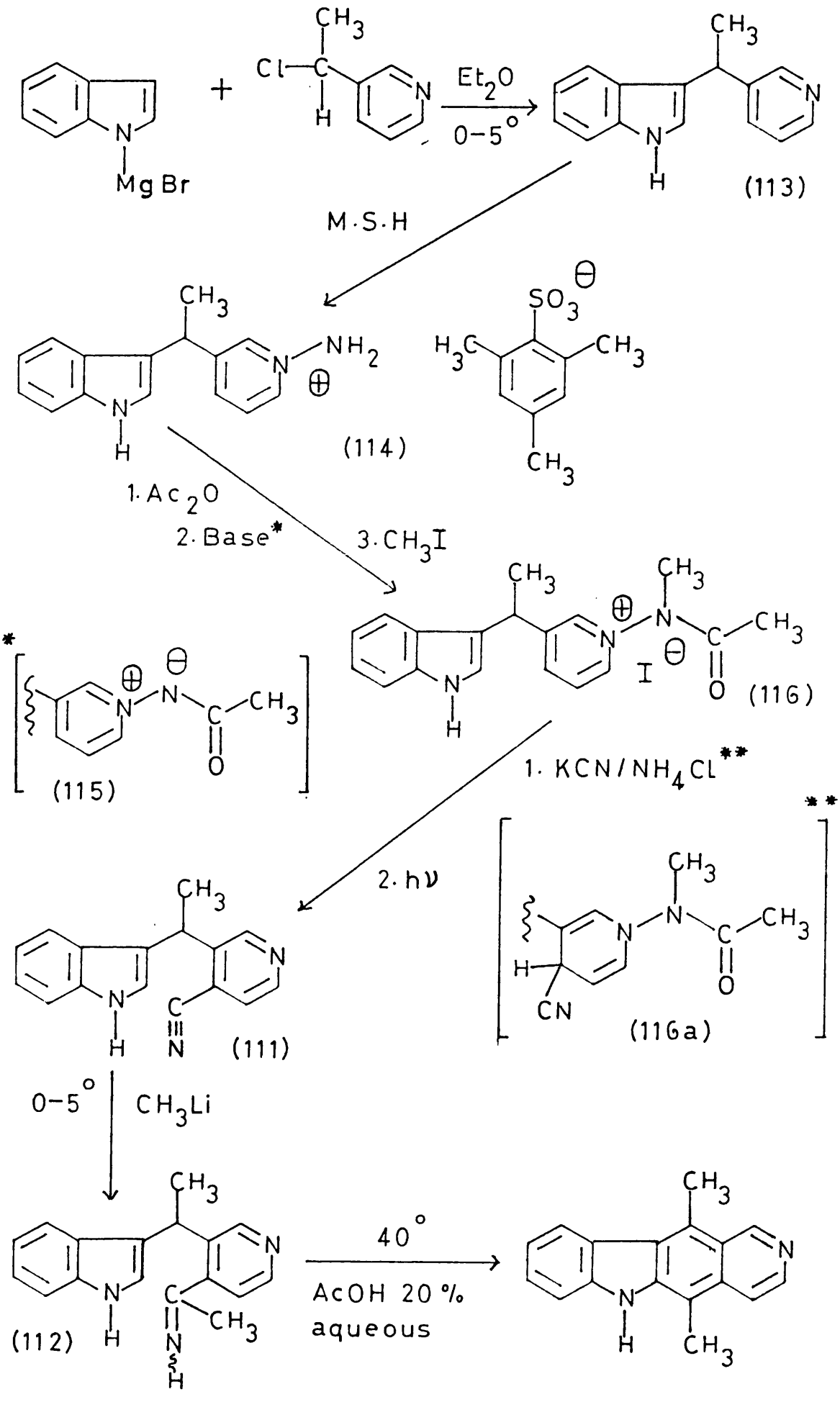
Ellipticine was obtained from this nitrile by a method pioneered in this laboratory[53, 54]. Reaction of (111) with four equivalents of methyl lithium gave the methyl imine (112), which was not isolated, but treated directly with 20% aqueous acetic acid at 100° to furnish a 80% yield of ellipticine.

This synthesis has potential versatility because either the α-metallated isonitrile used to generate the oxazole, or the dienophile component of the Diels-Alder reaction can be readily varied to give access to ellipticines with differently substituted D rings. However, the quoted yield of the Diels-Alder reaction is poor, (16%), and extensive purification is required. The yields of the individual stages are quoted in (Scheme 26) and the overall yield is of the order of 6%.

In conclusion it can be said that this novel method has potential for development but little practical utility in its present form.

In 1976 a potentially versatile synthesis of ellipticine was developed in this laboratory by Sainsbury and Schinazi[53, 54], who first prepared the carbonitrile (111), mentioned above in connection with Kozikowski's work[46]. This route employs very mild conditions throughout, and is shown in (Scheme 27).

Scheme 27


Et2O
0-5°
(113)
M.S.H
(114)
1. Ac2O
2. Base*
3. CH3I
(115)
(116)
1. KCN/NH4Cl**
2. hν
(116a)
(111)
0-5°
CH3Li
(112)
40°
AcOH 20 %
aqueous

The key reaction, that of a condensation between indolylmagnesium bromide and 3-(1-chloroethyl)pyridine in the cold, to give 3- 1-(3-pyridyl)ethyl indole (113) was based on the work of De Graw and his associates[55] who have carried out a number of similar reactions with encouraging results. Treatment of the condensation product (113) with mesitylene sulphonylhydroxylamine (M.S.H.) afforded the N-amino salt (114) which was acetylated with acetic anhydride to give the Zwitter ion (115). Methylation with hot methyl iodide furnished the methiodide (116), which was dissolved in water and treated with potassium cyanide in the presence of ammonium chloride to yield a 1,4-dihydrocyanopyridine intermediate (116a), which was not isolated but directly exposed to 'soft' ultraviolet light giving the carbonitrile (111). Attack upon the cyanide function by methyl lithium furnished the methyl imine (112). This was hydrolysed using 20% aqueous acetic acid at 40^{o} for ten minutes, thus effecting spontaneous ring-closure and aromatization to pure ellipticine in 60% overall yield from the indole (113).

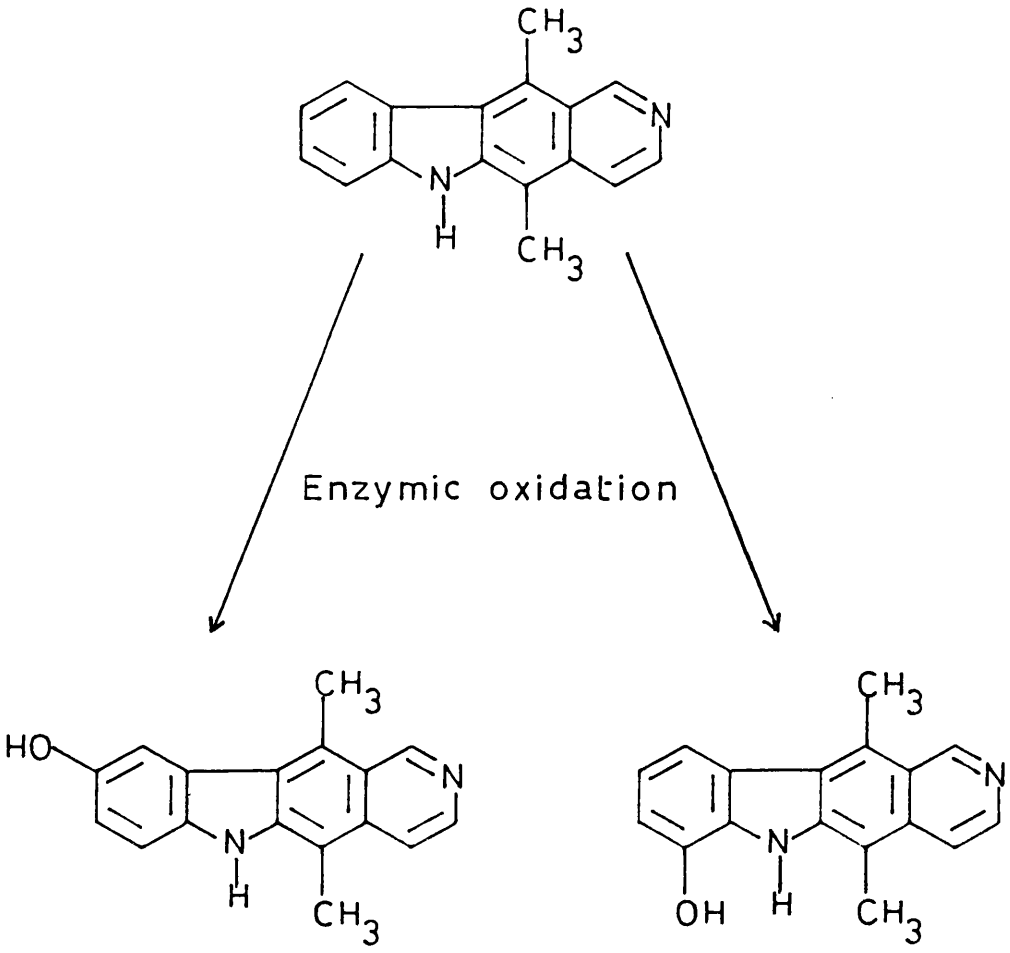
This route combines the advantages of a quick one step condensation and high productivity. At no stage are harsh conditions required, which is an advantage where thermo-labile or oxidation sensitive substituents are involved. Another practical advantage is that all the 4-substituted pyridine compounds synthesized, are isolable solids, rather than oils, as is the case for pyridines (35-41), used in the previous work carried out in our laboratory (see p. 23). The higher molecular weight of these indolylpyridylethanes must contribute towards this effect.

However, the Grignard condensation was not as efficient as the production of indoxylidines (see Scheme 7, p. 18), moreover 3-(1-chloroethyl)pyridine is, (as discussed on p. 114), somewhat unstable.

Nevertheless, the mildness of the conditions, and the potential for making a variety of ellipticines with various oxygenated substituents, made us look on this approach with optimism. We therefore chose this route as the most potentially promising to employ in our projected synthesis of 8-hydroxyellipticine, which forms the first part of the work to be described later in this thesis (see discussion section p. 112).

Recent studies concerned with the Metabolism of Ellipticine

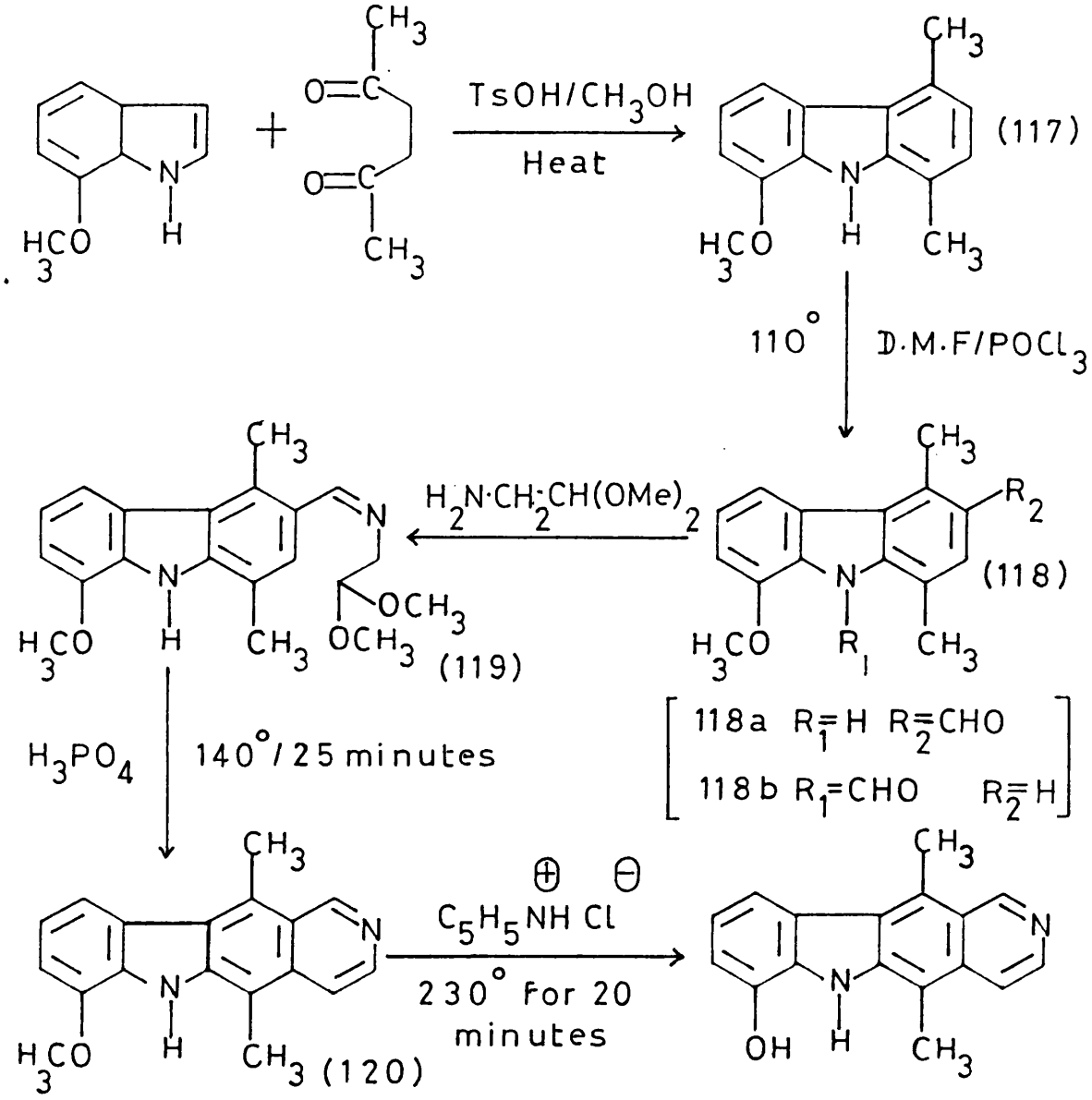
Some interesting work has been carried out on the metabolism of ellipticine in living systems in recent years. A notable example is the work of Lallemand and Mansuy[56], in Paris. These workers have investigated the metabolism of ellipticine in the rat, and have found that the parent molecule is oxidized by mono-oxygenase enzymes such as cytochrome P 450 in actively detoxifying hepatites. Thus the bile was examined for metabolic products, whereupon 9- and 7- hydroxyellipticines were identified. These were present as major and minor components respectively.



Major metabolite Minor metabolite

9-Hydroxyellipticine is a known compound,[57,58,59] hence it was identified by comparison of its H.P.L.C. retention time and spectral characteristics with an authentic sample. 7-Hydroxy-ellipticine however, was a new compound, and it was therefore necessary to achieve a synthesis of it, in order to positively identify this metabolite. These workers chose to use a modification of the synthesis of Cranwell and Saxton[8], (see p. 6), outlined in (Scheme 28).

Scheme 28.



7-Methoxyindole was prepared by the method of Kalir[60] and condensation with 2,5-hexadione gave the methoxy carbazole (117). The formylation method of Julia and Lallemand[61] was found to be superior to the original procedure, but these methoxylated substrates gave rise to two products (118a) and (118b). The product ratio was 9:1 and the isomers were separated by chromatography on silica gel using dichloromethane as the elutant. Pure (118a)

was condensed with 2,2-dimethoxyethylamine to give the imine intermediate (119) in 81% yield. Cyclization to 7-methoxyellipticine (120) was achieved by the action of hot 98% ortho-phosphoric acid. This compound was purified by vacuum sublimation followed by chromatography. The synthesis was completed by fusion of the pure 7-methoxyellipticine with pyridine hydrochloride at 230° according to the published method of Dat-Xuong[62]. This treatment O-demethylated the material to give 7-hydroxyellipticine, which was purified by chromatography.

The resulting material was identical in T.L.C. R_f values and spectral characteristics with the minor metabolite in the rat.

Recent work in the U.S.A. by Rosazza and Chien[63] has shown that microbial transformations of ellipticine with Aspergillus alliaceus yield 8- and 9-hydroxyellipticines. 8-Hydroxyellipticine was a new compound at this time, and its structure was determined largely by ^{1}H n.m.r. spectrometry at 90 MHz, in a comparative study with 9- and 7-hydroxyellipticines obtained by chemical O-demethylation of the corresponding methoxylated compounds by the published procedure mentioned above. The resulting ^{1}H n.m.r. data are shown in (Table 2).

Table 2.

H-Position	Ellipticine	9-Hydroxy	8-Hydroxy	7-Hydroxy
1	9.30	9.37	9.74	9.70
3	7.83(d,6Hz)	7.78(d,6Hz)	7.86(d,6Hz)	7.92(d,5.5Hz)
4	8.37(d,6Hz)	8.30(d,6Hz)	8.38(d,6Hz)	8.45(d,5.5Hz)
5-CH_3	3.23	3.20	3.17	3.26
6(NH)	10.93	10.65	11.12	11.50
7	7.37-7.60m	7.28(d,8Hz)	6.94(d,2Hz)	
8	7.37-7.60m	6.95(dd 8Hz and 2Hz)		7.06(dd 8Hz and 0.8Hz)
9	7.37-7.60m		6.71(dd 8Hz and 2Hz)	7.12(t,8Hz)
10	8.23m	7.70(d,2Hz)	8 15(d,8Hz)	7.86(dd 8Hz and 0.8Hz)
11-CH_3	2.77	2.73	2.75	2.96

Solvent, all D.M.S.O. d_6, all δ p.p.m. and J values.60 MHz (Ellipticine and 9-hydroxy), 90 MHz (8-hydroxy), 250 MHz (7-hydroxy).

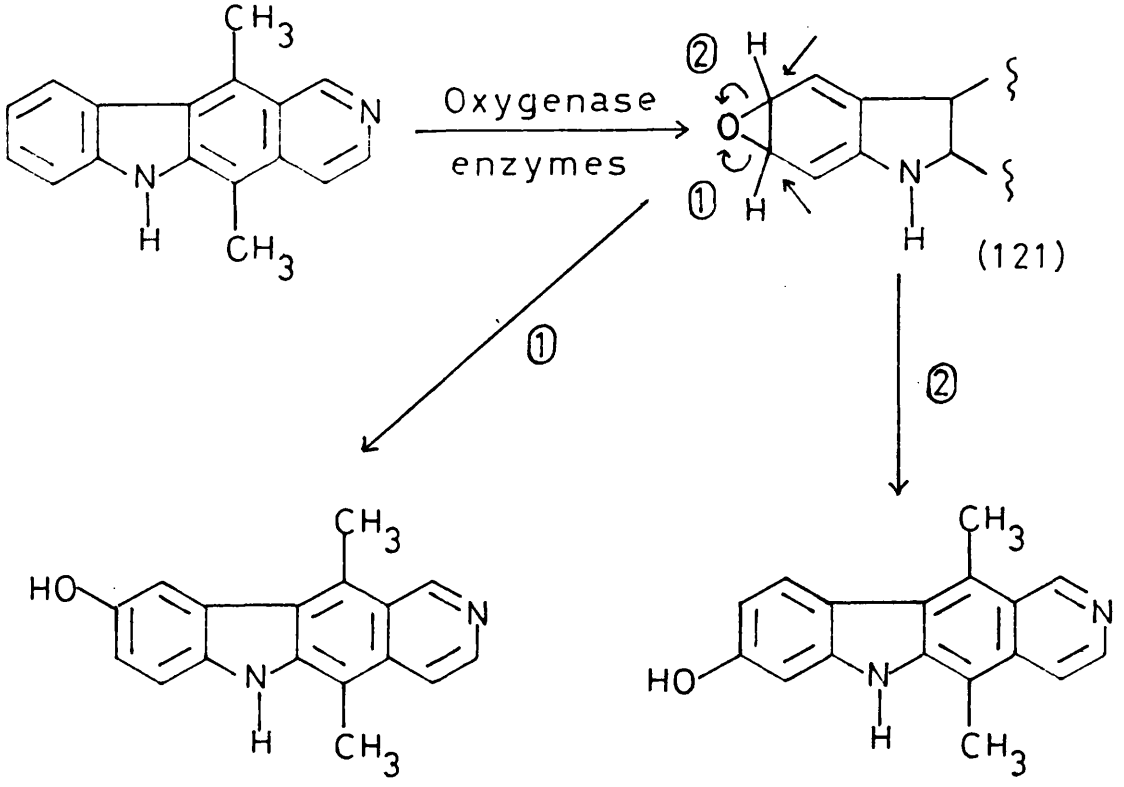
It can be seen from this data that the 8-hydroxyellipticine isolated by Rosazza and Chien exhibits the expected A.B.X. pattern, which readily distinguishes it from unsubstituted ellipticine and the 7-hydroxy compound. The A.B.X. patterns of the 8- and 9-hydroxy isomers differ only slightly, but a comparison of the

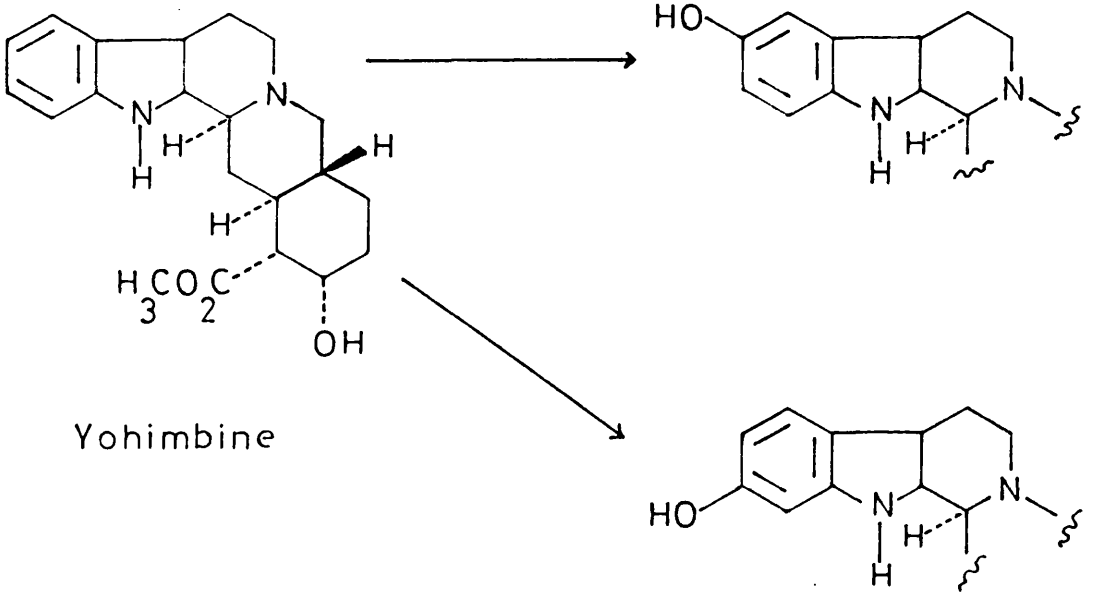
1OH signals shows a significant (0.45 p.p.m.) downfield shift for the 8-OH compound with respect to 9-hydroxyellipticine. This presumably reflects a degree of electron deficiency at the expected position, meta- to the 8-OH group.

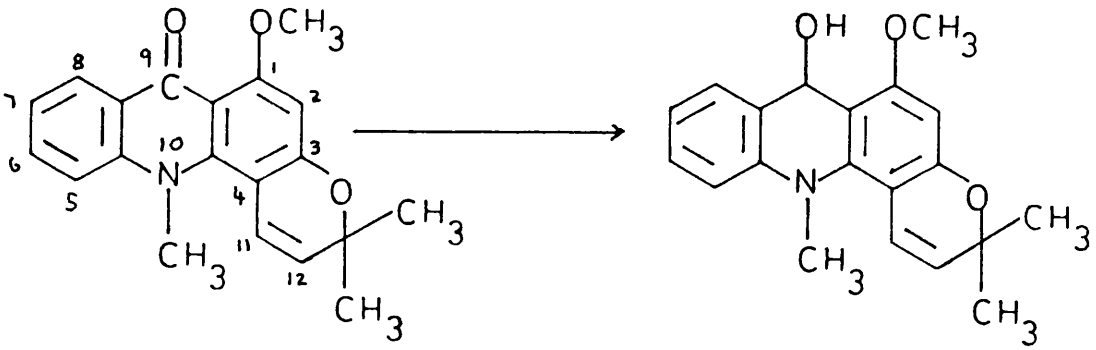
Other physical data including the melting point and T.L.C. analysis in three different solvent systems, served to distinguish the new compound as 8-hydroxyellipticine.

Microbial hydroxylation is normally considered to occur according to the usual rules of electrophilic aromatic substitution. Thus, the isolation of 9-hydroxyellipticine as a major metabolite is to be expected. More fascinating, however, is the observed production of both 8- and 9-hydroxyellipticines by the same microbial culture. Rosazza[63], speculates that both metabolites arise through an unstable, common intermediate, the 8,9-arene oxide (121), shown in (Scheme 29). This intermediate will then provide either of these metabolites depending upon the manner of epoxide opening. Precedence exists for this reaction in the hydroxylation of the equivalent C-10 or C-11 positions of yohimbine to give either 10 or 11-hydroxyyohimbines in Cunninghamella species[64], and other fungi give 9-hydroxyacronycine[65], which is the major mammalian metabolite of this anti-tumour alkaloid.

Scheme 29.



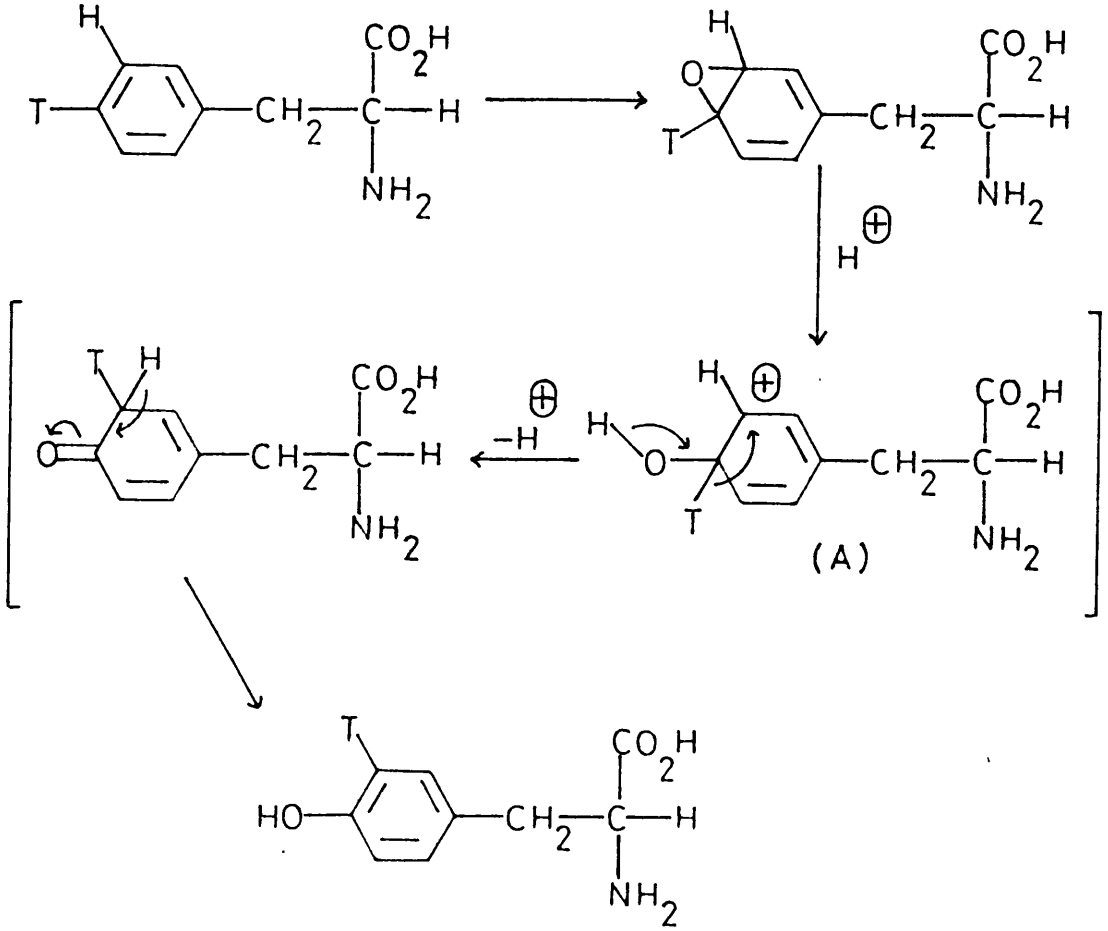




Acronycine

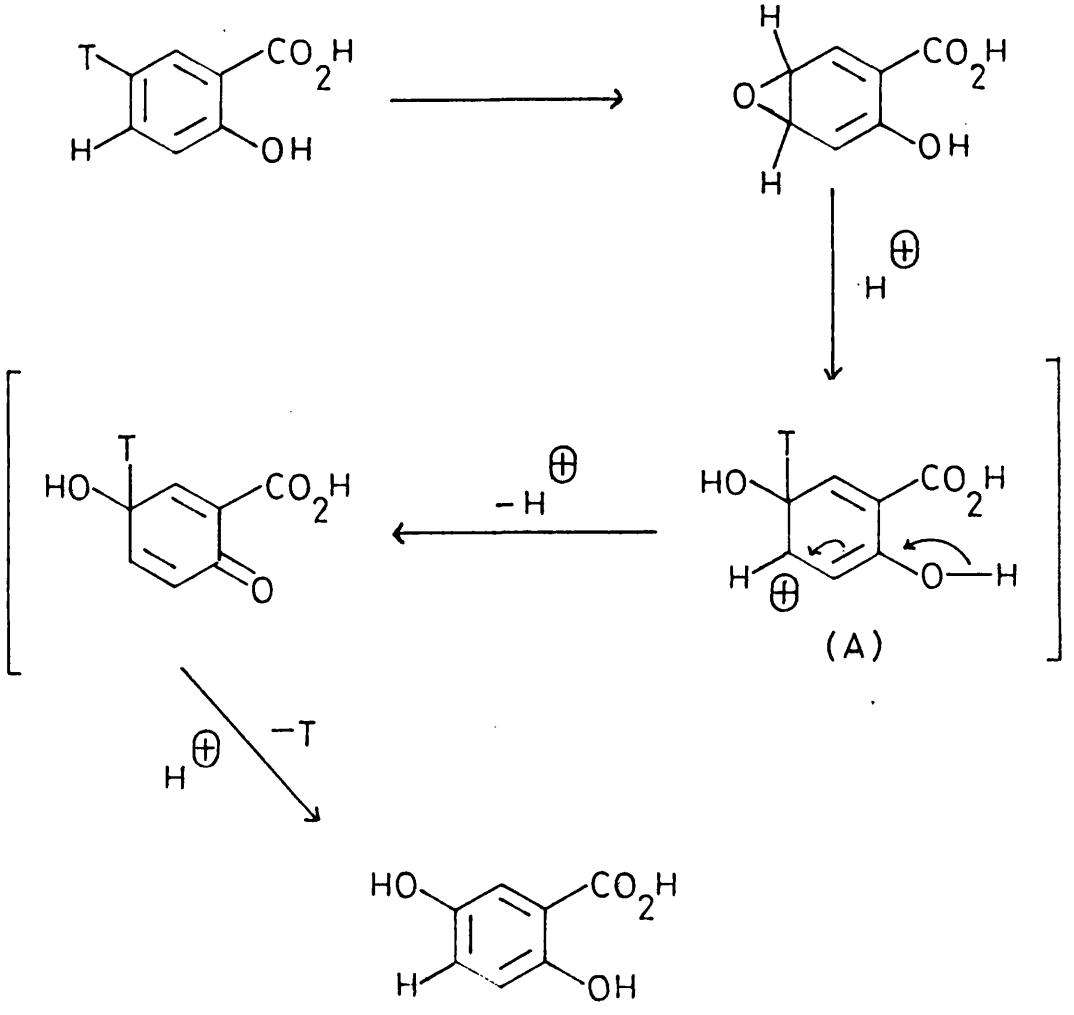
Hydroxylation of aromatic substrates by many fungi occurs through the formation of arene oxides of the type shown in (Scheme 29)[66], and it is interesting to speculate on the possibility of the N.I.H. shift phenomena occurring in this reaction. This process, which has been shown to occur in many fungi as well as mammals is illustrated in (Scheme 30) for tritium labelled phenylalanine[67].

Scheme 30.



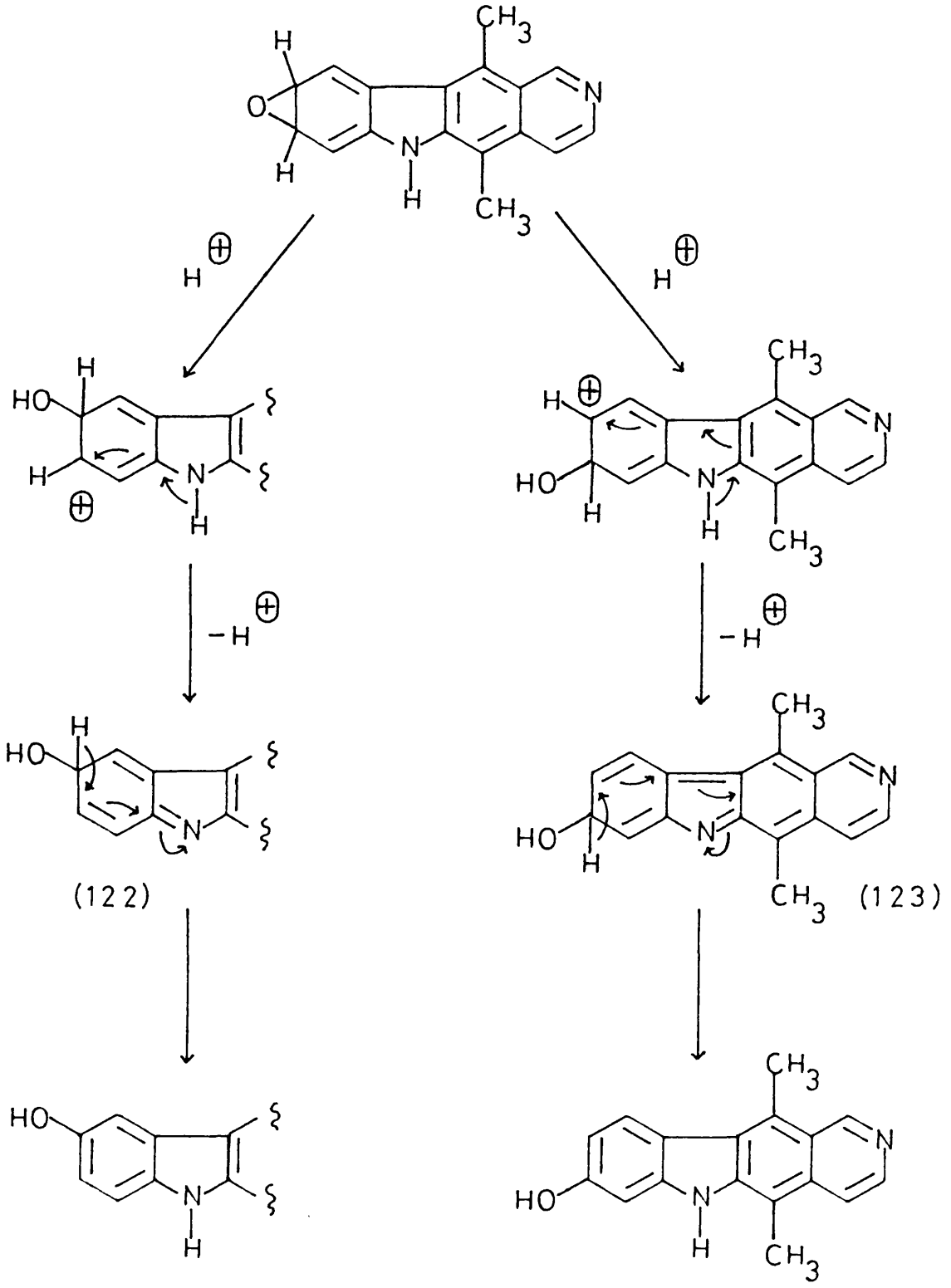
This conversion involves the 1,2-shift of a hydride ion or the anion of another substituent should one be located at the hydroxylated carbon atom. This only takes place when the compounds shown as (A) in (Schemes 30 and 31) cannot stabilize their positive charges by a shift of electrons involving a second electronegative atom. Thus, no N.I.H. shift occurs in the formation of 2,5-dihydroxybenzoic acid (Scheme 31). Bond strengths at the site of hydroxylation decide which substituent will be lost, in this case C-OH is stronger than C-T, so the tritium atom is eliminated.

Scheme 31.



It can be seen from the above argument that the N.I.H. shift is unlikely to be observed in the case of ellipticine hydroxylation because of the possibility of charge delocalization with the ring 'B' nitrogen atom occurring as outlined in (Scheme 32).

Scheme 32.



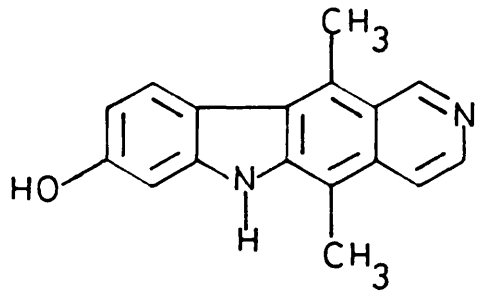
However, if the intermediate (123) is formed it is likely to be of higher energy than its C-9 substituted counterpart (122). It would form an interesting study to prepare ellipticine with a tritium atom at the C-8 position (from an appropriately substituted indole), subject this to microbial hydroxylation and test the products for tritium inclusion at the C-9 position. This would show whether a N.I.H. shift does occur during 8-hydroxylation.

This is the first reported work on the microbial metabolism of ellipticine, and 8-hydroxylation is a hitherto unknown metabolic transformation. Due to the fact that 9-hydroxyellipticine has the highest degree of antineoplastic activity of any known ellipticine[68], this new derivative generated some interest, but the 8-hydroxy compound required extensive chromatographic separation from its isomer and insufficient pure material was obtained to carry out a full pharmacological evaluation.

Clearly there was a need for a chemical synthesis to make larger quantities available. Firstly to establish beyond doubt the validity of the spectroscopic assignment and to decide its clinical potential. At the time this paper appeared we were actively engaged in designing a synthetic route to this compound, and much of the work that follows in this thesis is concerned with realizing this aim.

DISCUSSION

Our first objective was the preparation of sufficient 8-hydroxyellipticine (124) for biological testing.

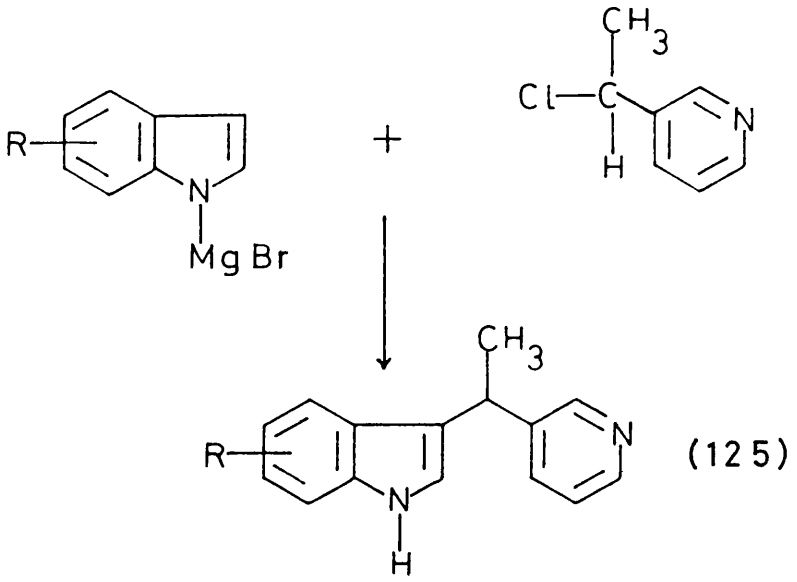


(124)

As mentioned previously (p. 74), interest in this new derivative arose as a natural consequence of the very encouraging results obtained with 9-hydroxyellipticine against some forms of human leukaemia.[68] 8-Alkoxyellipticines were also required in order to assist in the testing of pharmacological theories connected with their in vivo reactions (see p. 221 for a discussion of ellipticine pharmacology).

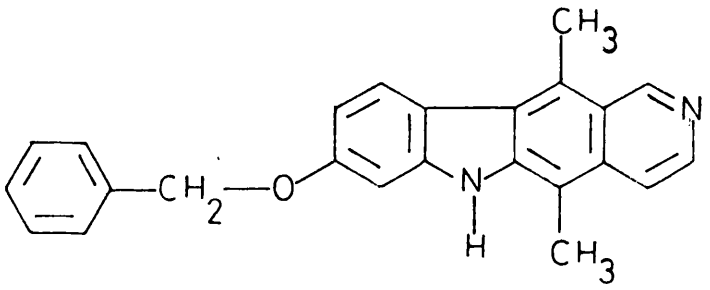
Thus, we decided to synthesise this derivative by a modification of the successful procedure previously employed in this laboratory by Schinazi[54] and Driver[69] to prepare ellipticine itself and various alkyl derivatives, (see Scheme 27, p. 62).

The key reaction of this synthetic route is outlined below.



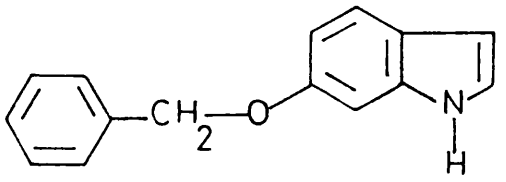
(125)

The appropriate indolylmagnesium bromide is condensed with 3-(1-chloroethyl)pyridine to give an indolylpyridylethane of the type (125). Applied to the synthesis of 8-hydroxyellipticine this requires the preparation of a protected 6-hydroxyindole, and the O-alkyl or O-aryl ethers were obvious choices because they are reasonably stable and the resulting alkoxy or benzyloxy ellipticines would be interesting compounds themselves. We also wished to culminate the synthesis with a precursor which could be converted easily into 8-hydroxyellipticine under mild conditions. Thus, our initial target was 8-benzyloxyellipticine (126), since it is known[70], that the benzyloxy function is easily cleaved by catalytic hydrogenolysis.[71,72]



(126)

The first requirement was, therefore, to prepare a stock of 6-benzyloxyindole (127).

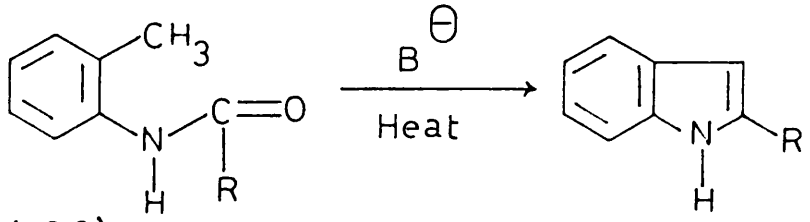


(127)

Of the routes to indoles not substituted at the 2- or 3-positions,[73] and known to give reasonably good yields, we considered the following.

(A) The Madelung Synthesis

This involves the very vigorous base treatment of an ortho-toluidide.



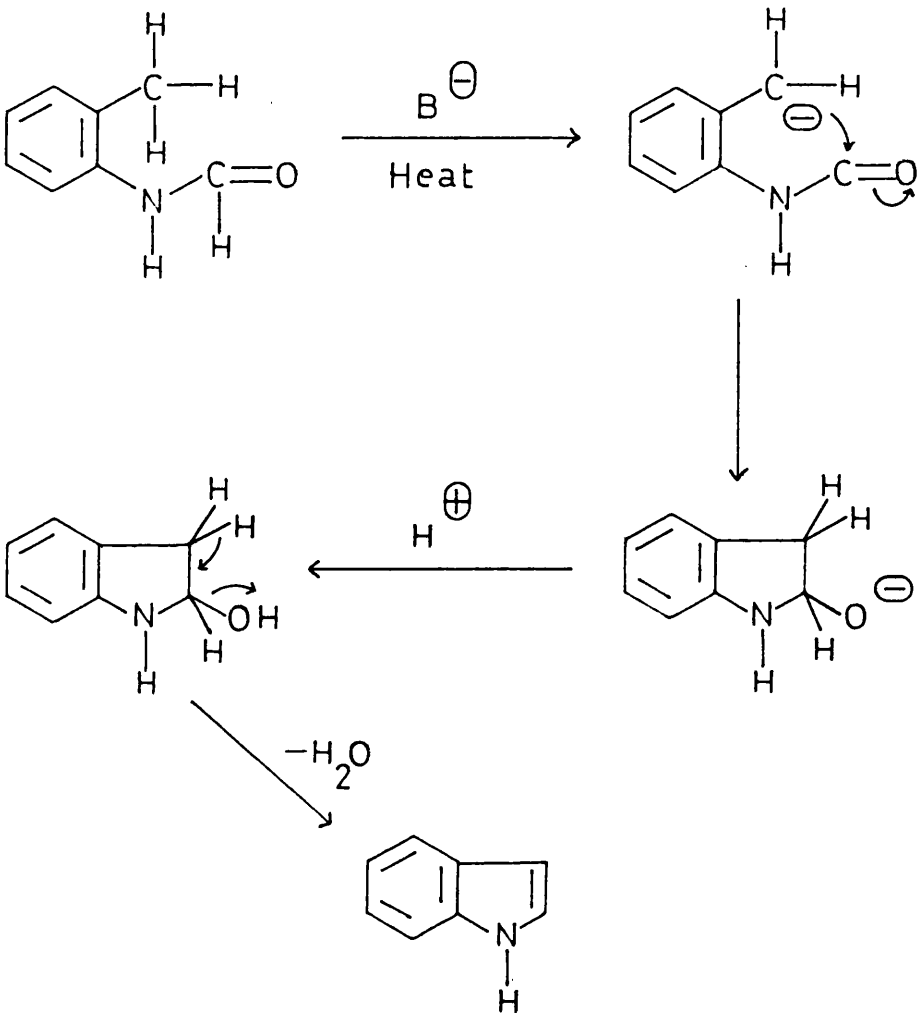
(128)

This synthesis is normally used to prepare indoles substituted in the 2-position with alkyl or aryl groups, however, Tyson[74], obtained indole in this way, by heating the formamide (128; R = H) with potassium tertiary butoxide at 350-360°.

The full mechanism of the Madelung synthesis is not known with certainty[75], but it is usually assumed to proceed by base removal of a proton from the 2-methyl group affording a carbanion which attacks the carbonyl function in an intramolecular reaction, protonation and elimination of water then give the product indole as outlined in (Scheme 33).

Successful cyclization depends on very severe conditions and it is interesting to consider possible reasons for this. Firstly, the hydrogen atoms of the methyl group in the substrate (128) are not particularly 'acidic' and require a strong base and a high temperature for their removal. Secondly, one may speculate that under these conditions initial deprotonation occurs from the nitrogen atom, and as this would require the generation and subsequent reaction of a dianion it would be a very high energy process.

Scheme 33

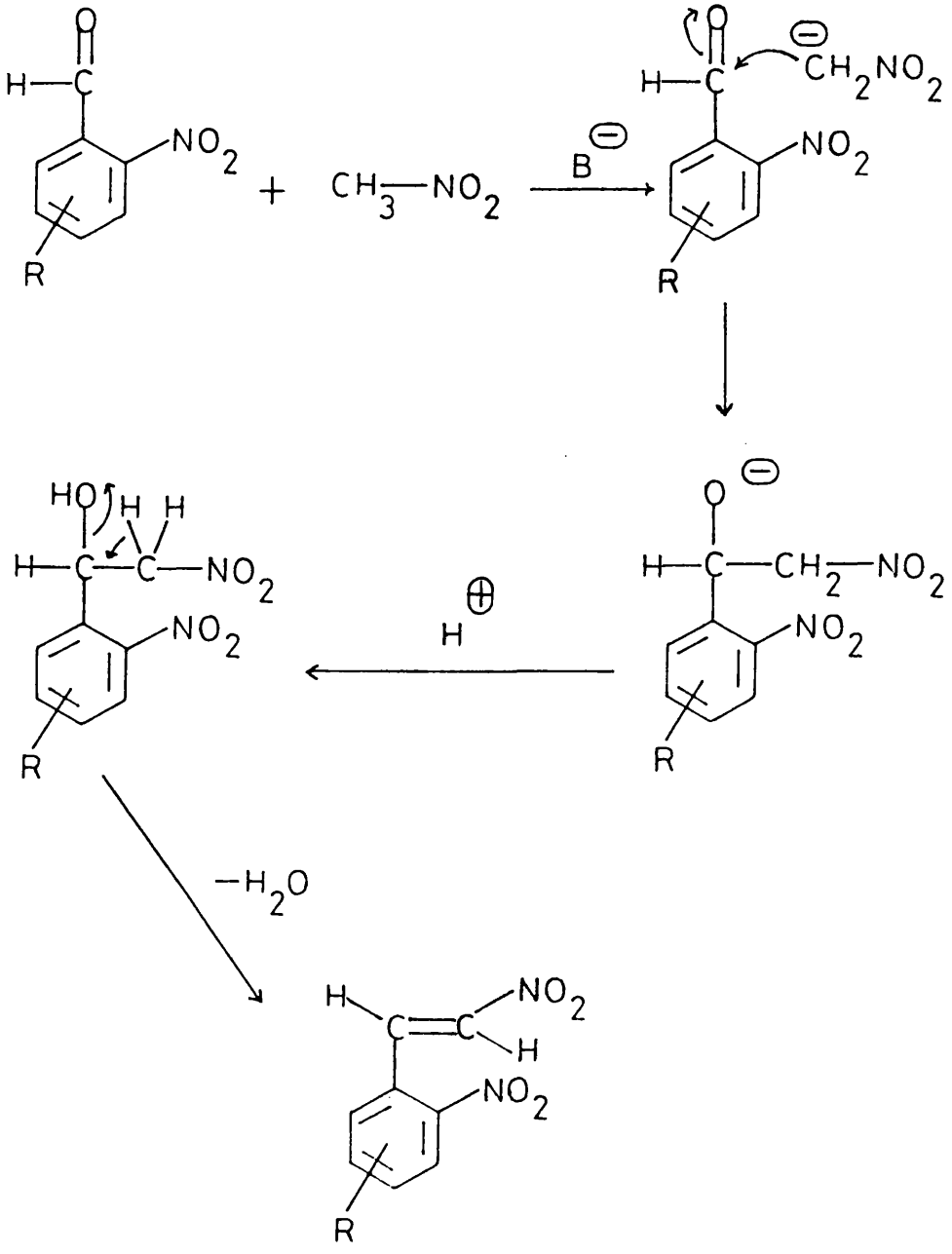


In either case, temperatures in excess of 300° and strong bases such as potassium tertiary butoxide or sodamide are obvious disadvantages as this treatment is known[76] to cause a high proportion of the substrate to undergo decarbonylation, and large quantities of 2-toluidine have always to be separated from the product. Hence high yields are rare in this synthesis.

(B) The reductive cyclization of 2,β-dinitrostyrenes

This method involves the condensation of a 2-nitrobenzaldehyde with nitromethane under basic conditions to give the appropriate 2,β-dinitrostyrene as outlined in (Scheme 34). These compounds can be reductively cyclized to indoles, and the reductive step is most conveniently carried out by catalytic hydrogenation,[77] or alternatively by electron transfer reagents such as iron and acetic acid.[78]

Scheme 34

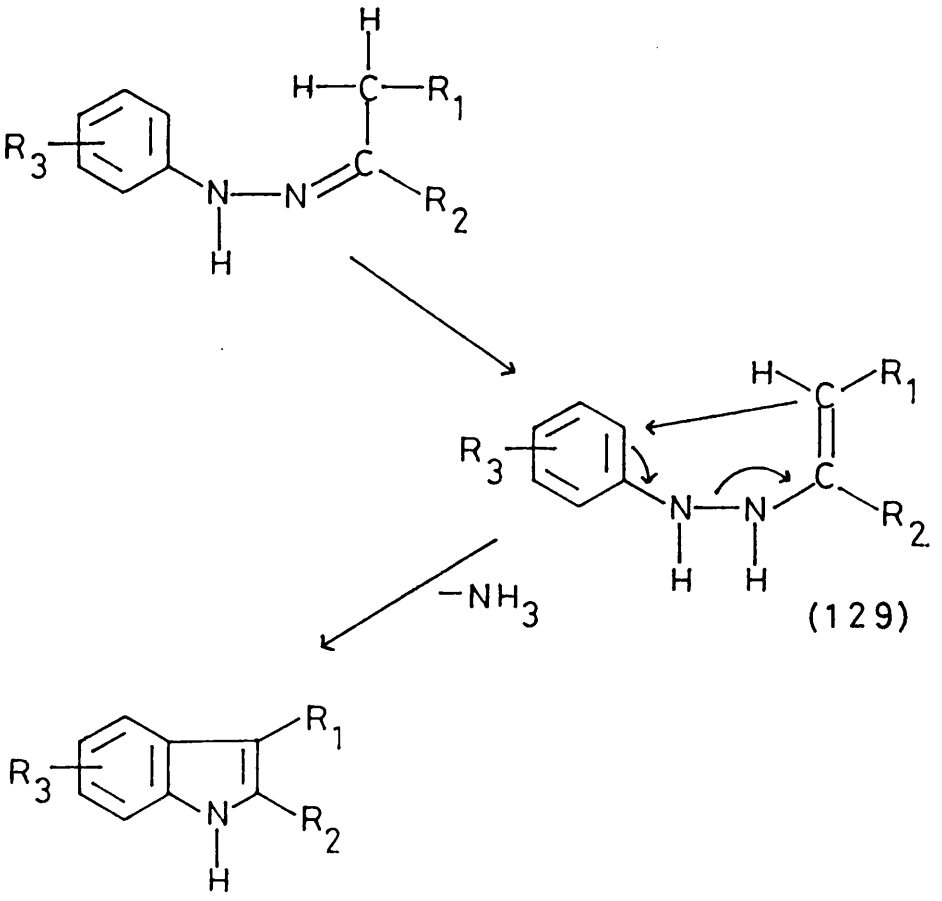


Generally speaking this is a short, high yielding synthesis but its usefulness is dependent upon the availability of the appropriate 2-nitrobenzaldehyde.

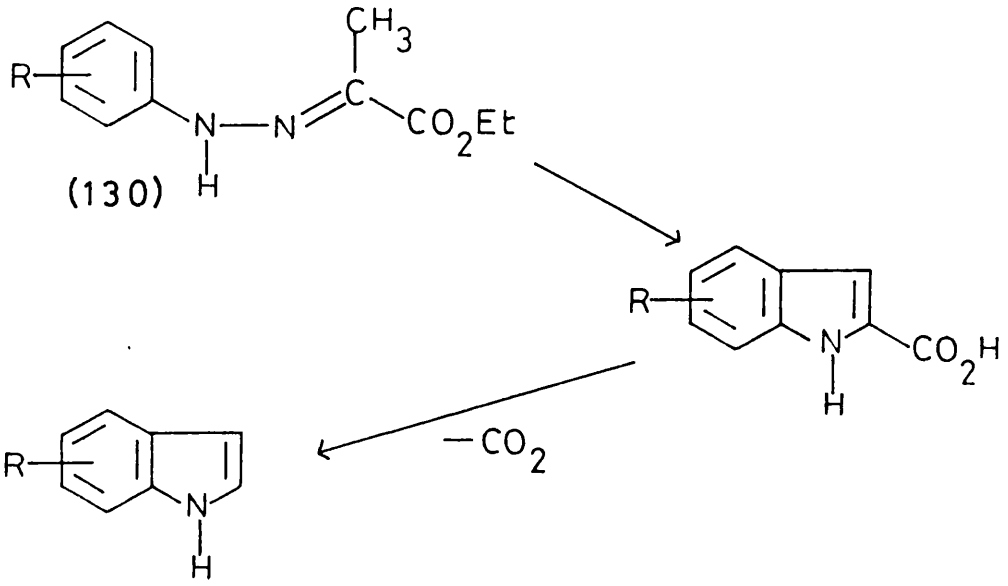
(C) The Fischer Synthesis

This method normally involves heating a phenylhydrazone with acids or Lewis acid catalysts[79], but in many cases catalysts are not required and it is sufficient to simply heat the phenylhydrazone in an inert solvent[80]. The key step is the formation of a tautomer (129), of the original hydrazone which then undergoes a [3,3] sigmatropic shift[81]. Ammonia is lost and an indole is formed, as outlined in (Scheme 35).

Scheme 35



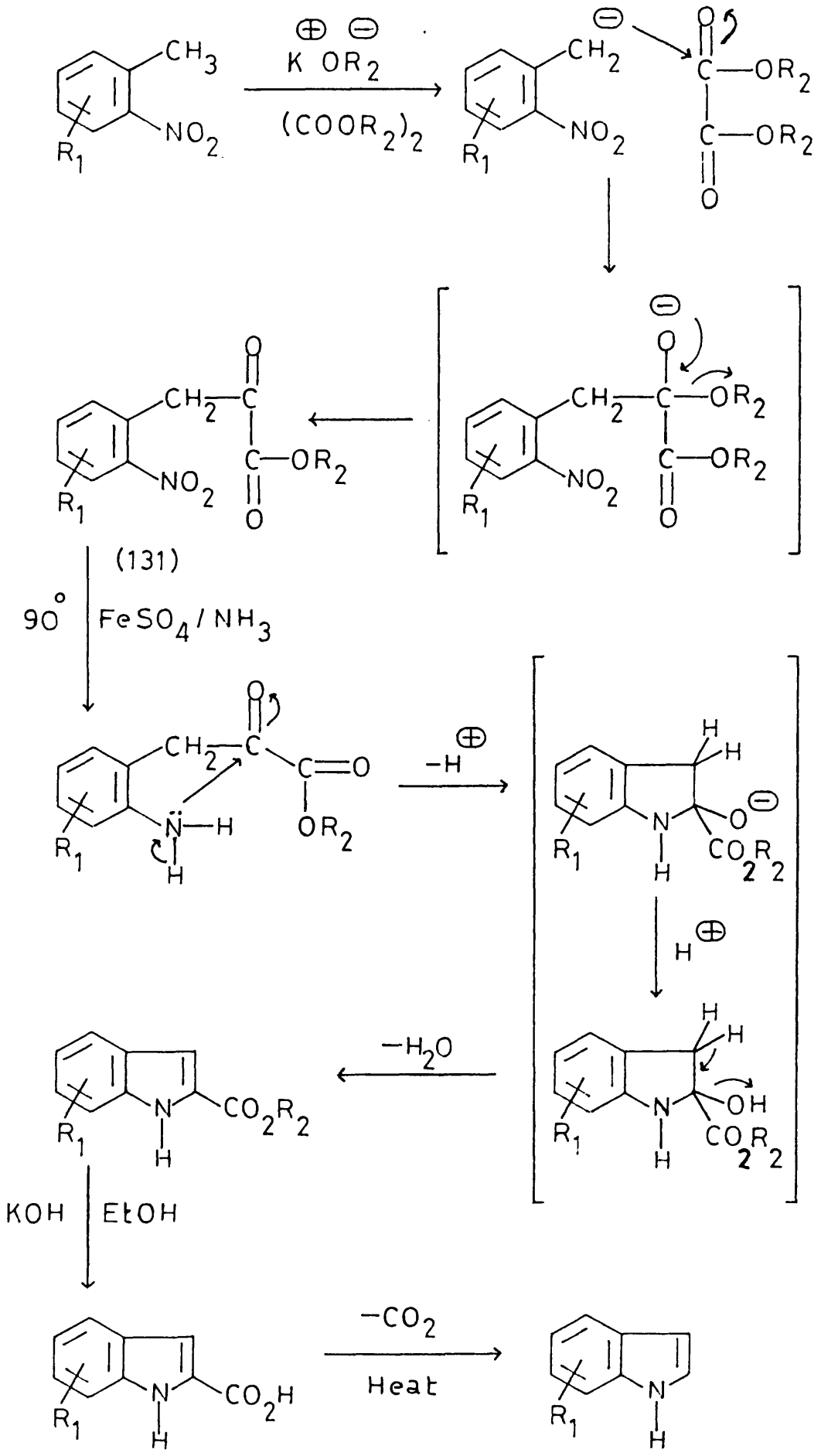
The widest use of this reaction is to make 2- and 3-substituted indoles, but compounds unsubstituted at these positions can be prepared by cyclizing pyruvic acid phenylhydrazones or their esters (130).[82] This gives the corresponding indole-2-carboxylic acid which can be subsequently decarboxylated as shown below.



(D) The Reissert Synthesis

The Reissert synthesis involves the base catalysed condensation of a 2-nitrotoluene with a dialkyloxalate ester to give an intermediate of the type (131), which on reduction cyclizes to an indole-2-carboxylate ester. Finally hydrolysis and decarboxylation yield the desired indole,[83,84] as outlined in (Scheme 36).

Scheme 36

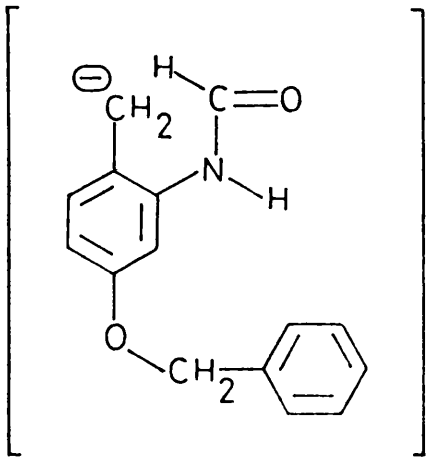


Here, once again it is clear that the general applicability of the synthesis is governed by the availability of the appropriate 2-nitrotoluene. On considering the application of the above methods to our own particular substrate, we came to the following conclusions.

(A) The Madelung Reaction

We did not consider this method to offer a satisfactory synthesis in our particular case, because, as indicated above[74], it involves very harsh conditions. In practice, such techniques often give rise to much charring of the substrate and product, with consequential purification difficulties. We also foresaw problems with the formation of the intermediate (132), since it is relatively destabilized by the presence of a benzyloxy group in the para-position.

(132)

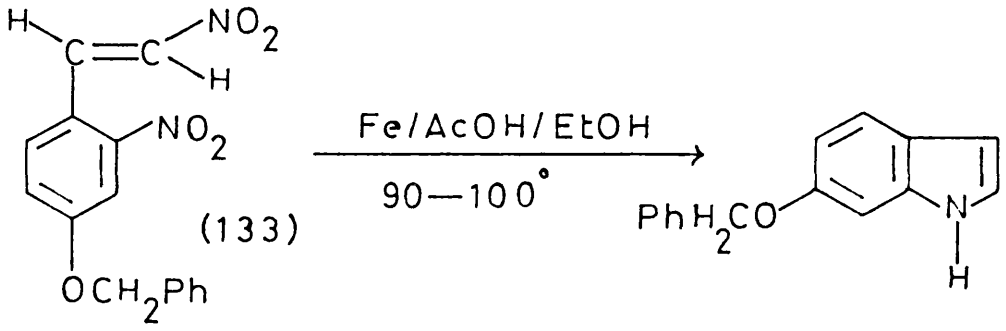


We therefore expected that conditions for a Madelung cyclization of this type would be at least as severe as for the unsubstituted analogues, and bearing in mind that a benzyloxy group is not difficult to cleave we decided to discount the use of this approach.

(B) The reductive ring closure of 2,β-dinitrostyrenes

We could not apply this otherwise useful method directly, because catalytic hydrogenolysis of the benzyl group would occur, to give 6-hydroxyindole. This would present a problem because it would need to be selectively re-O-benzylated before the remaining steps in the synthesis could be pursued.

Acid catalysed ring closure of the 2,β-dinitrostyrene (133) was also considered, using iron and ethanolic acetic acid at 90-100°.

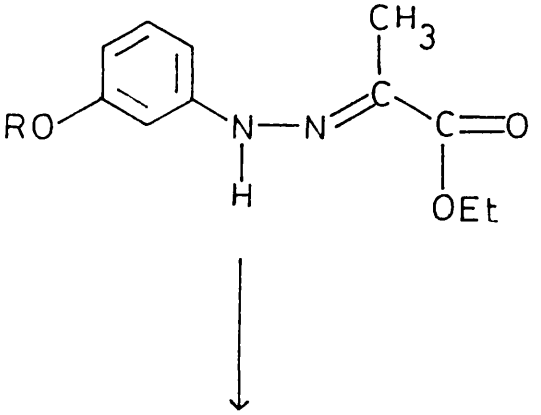


Suvorov and co-workers[85] have reported a synthesis of 6-benzyloxyindole by this method. However, McOmie[70] reports that benzyl phenyl ether systems are easily cleaved in acidic media, and indoles themselves are not very stable under acid conditions,[86] particularly at elevated temperatures. It is also to be expected that the extra electron availability present in alkoxy or benzyloxyindoles will increase their sensitivity to acid catalysed polymerisation and oxidative degradation.

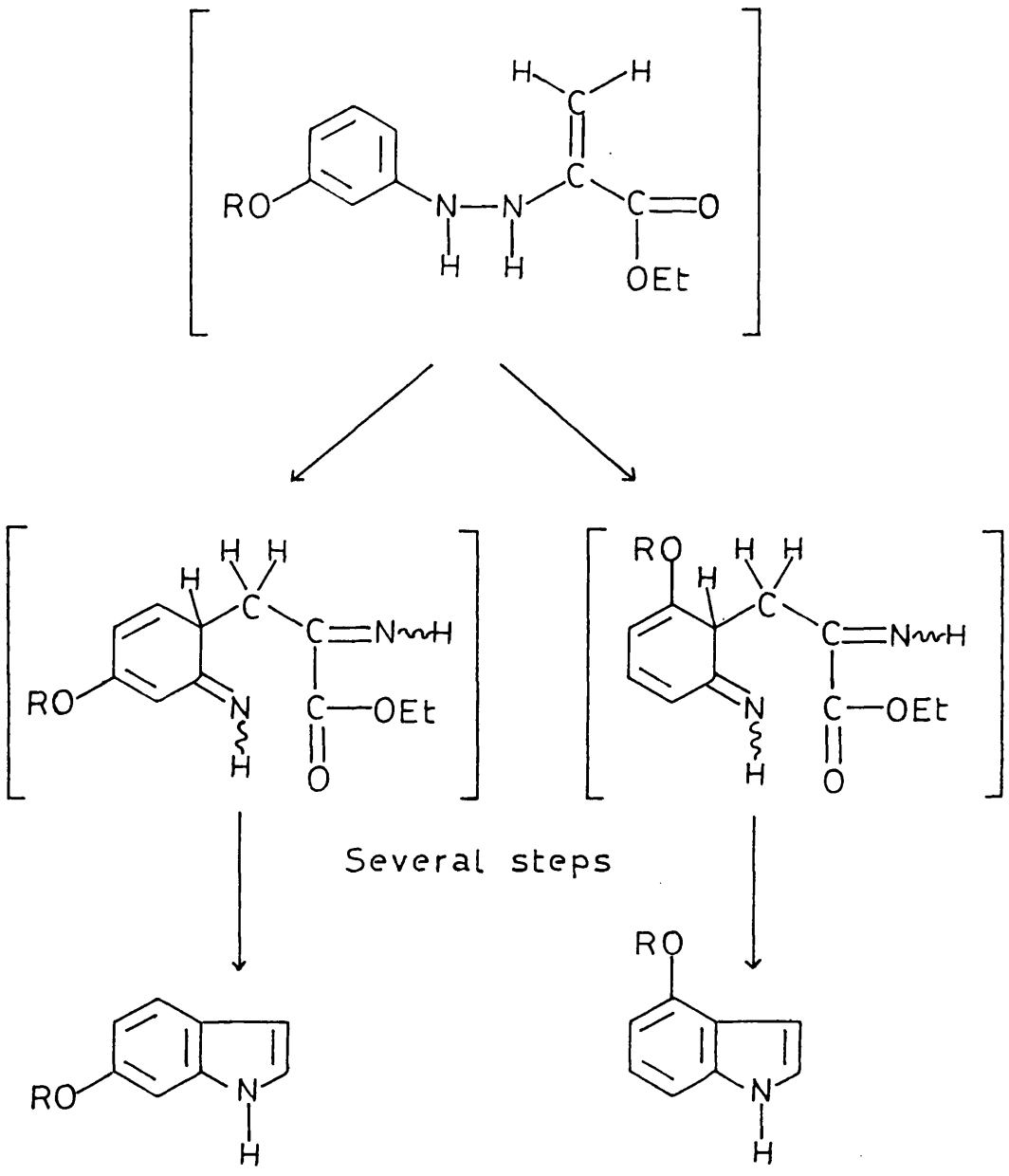
Suvorov comments that 6-benzyloxyindole is sensitive to both acids and heat. Furthermore Ek and Witkop,[78] found that extensive purification procedures were required to obtain 5- and 7-benzyloxyindoles by this method, and Benigni and Minnis[87] achieved low yields of 5,6-dibenzyloxyindoles by the use of a similar procedure. Spande[88] also comments that catalytic reduction of 2,β-dinitrostyrenes is more popular than the chemical methods in modern practice. For these reasons, we felt that this method was unlikely to be of practical value where (10-20g) quantities of the indole were required, thus we rejected this approach.

(C) The Fischer Synthesis

We realised that an unambiguous synthesis of 6-benzyloxyindole was mandatory, and that the traditional Fischer method does not fulfil this requirement. The ring closure of alkoxyphenylpyruvate hydrazones of type (134) give rise to both 4- and 6-substituted products because of alternative ring closures as shown.



(134)

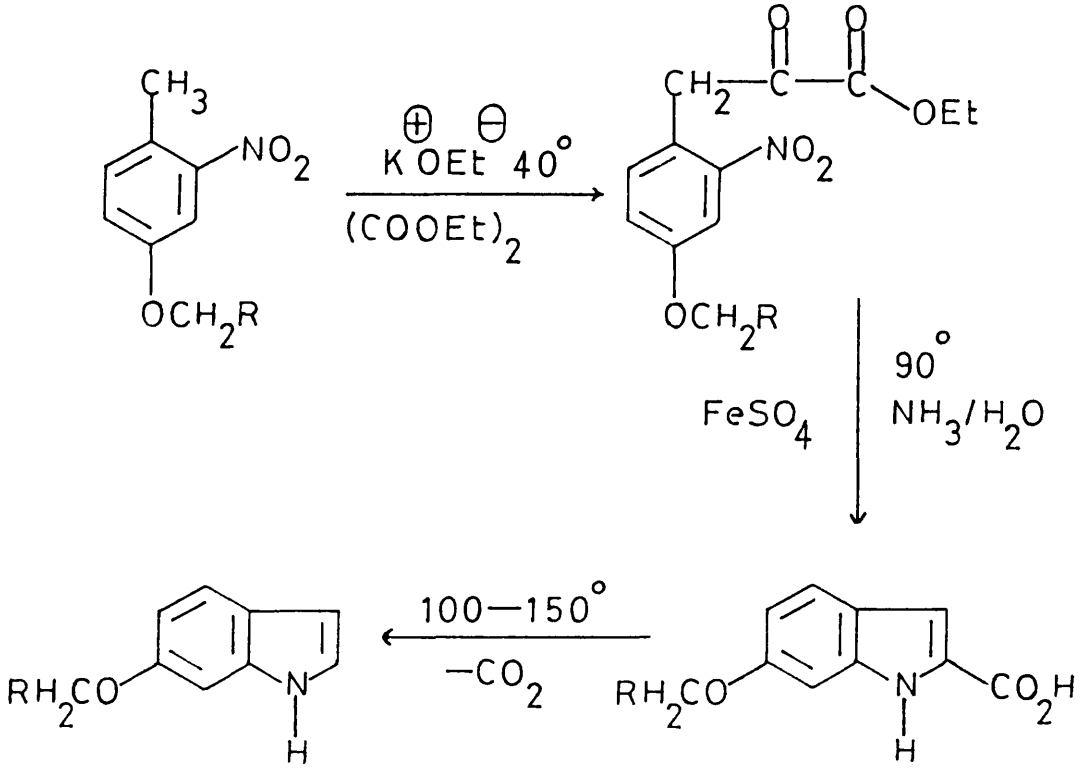


One may argue that stereochemical constraints in hydrazones of the type (134), might well lead to a weighting of the product ratio in favour of the desired isomer. However, previous experience has shown that it is difficult to separate indole isomers of this type on a preparative scale, particularly since we did not have an efficient high pressure liquid chromatograph at this time.

In view of these difficulties, the Reissert Synthesis was selected as the most promising route to pursue. A literature survey of this method revealed that Blaike and Perkin[83], Burton and Stoves[89] and Robertson *et al*[90], have prepared hydroxyindoles in this way *via* their benzyloxy precursors; although the overall yields were only in the region of 20%. This, we felt, was due to the same problem as cited previously for the Madelung reaction, that of decreased 'acidity' of the methyl protons in these *para*-oxygenated derivatives, since 2-nitrotoluene[91] and its alkyl derivatives react with ease in the Reissert Synthesis.

However, this method has the advantage that all the steps up to the final decarboxylation can be conducted at temperatures of less than 100°, under relatively mild conditions that do not require acidic media, as outlined in (Scheme 37).

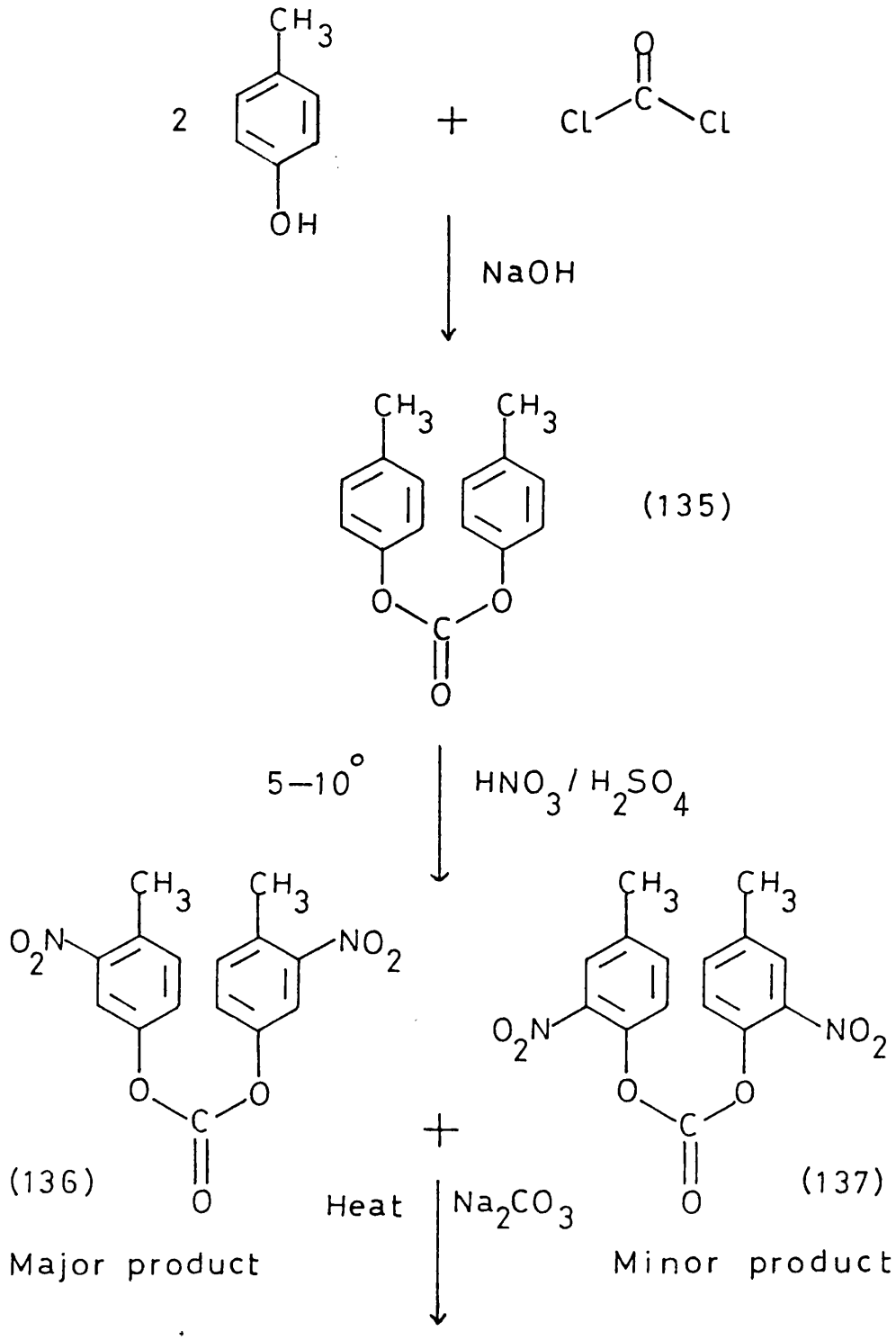
Scheme 37

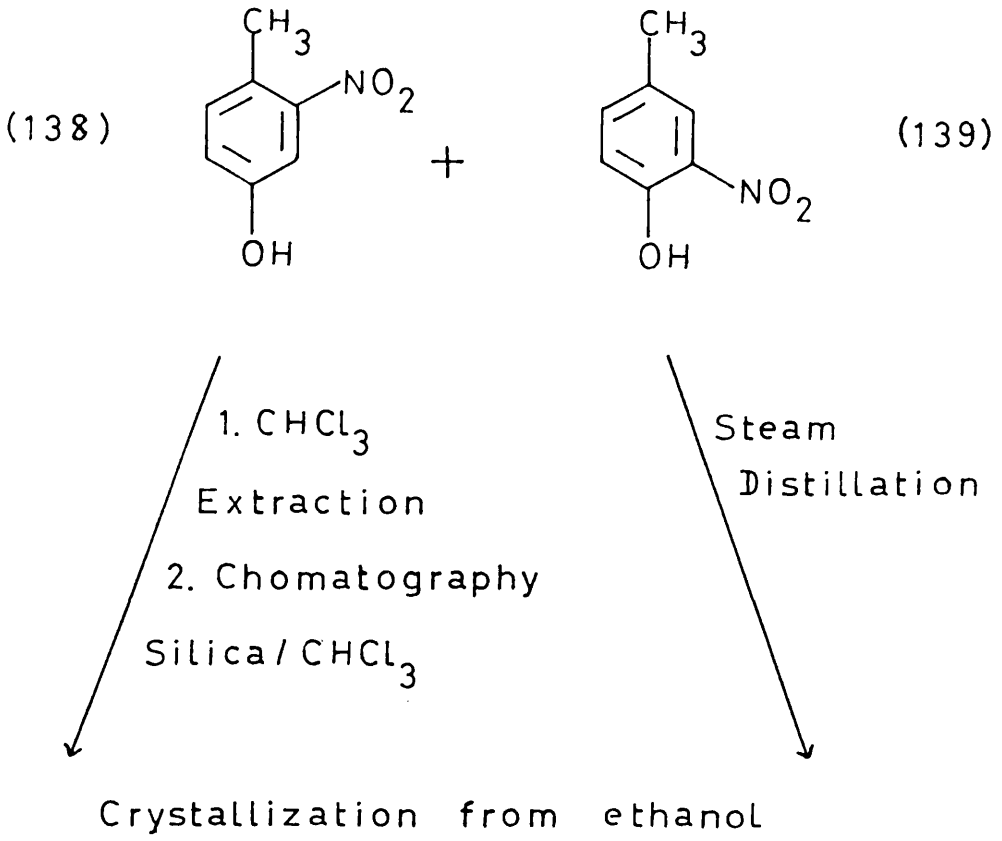


Where, R = C_6H_5 or CH_3

Blaikie and Perkin[83] began their synthesis with 4-methyl-3-nitrophenol (138). This compound is commercially available, but very expensive (£362 for 250g) at the time of writing. Thus, we undertook to prepare it, using the method of Copisarow[92], as outlined in (Scheme 38).

Scheme 38.

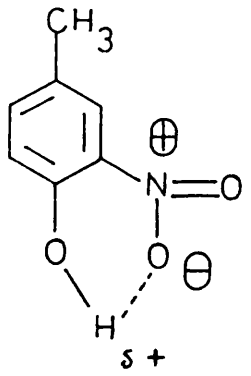




Condensation of 4-cresol with carbonyl chloride (phosgene) was achieved in a rapidly stirring mixture of toluene and aqueous sodium hydroxide, to give 4-tolylcarbonate (135) in 62% yield. This was nitrated with a mixture of concentrated sulphuric and nitric acids in a ratio of 2:1 at 5-10°, to give a mixture of the 3- and 2-mononitro-4-tolylcarbonates (136) and (137), in a ratio of 3:2. The weighting of the product ratio in favour of the desired compound (136) is to be expected as there is a certain amount of steric hindrance at the benzene positions ortho- to the bulky carbonate function.

Hydrolysis with hot aqueous sodium carbonate solution liberated the corresponding isomeric nitro phenols (138) and (139). These compounds were separated by steam distillation, the ortho-

isomer (139) being more volatile because it exists as an intramolecular hydrogen bonded chelate (139a).

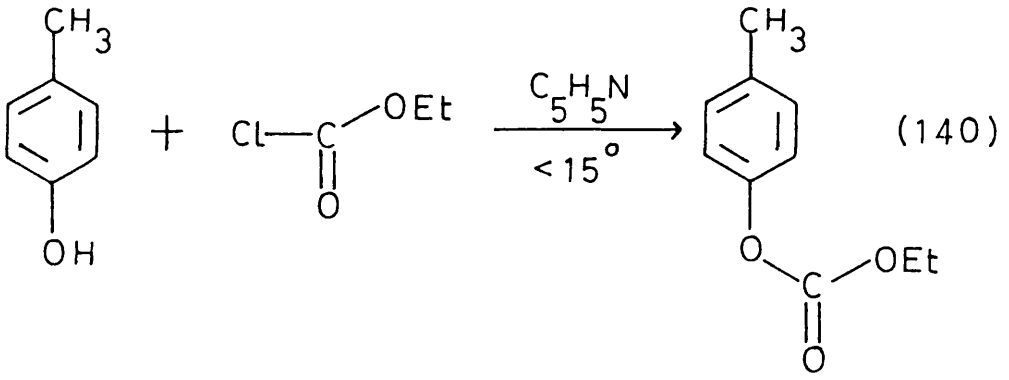


(139a)

Unfortunately this treatment left the desired compound (138) in a rather impure form that required chromatography on silica gel using chloroform as the elutant. This gave 4-methyl-3-nitrophenol as an orange amorphous solid which crystallized from ethanol as shining amber plates.

This route gave an overall yield of 40% after purification, and in order to build up a stock of 4-methyl-3-nitrophenol it would have been necessary to repeat this sequence a number of times.

As an alternative, and to avoid the toxicity and unpleasant handling characteristics of phosgene we decided to prepare 4-toluene ethylcarbonate (140), and nitrate it as before. This compound was simply prepared[93] by the condensation of 4-cresol with ethylchloroformate in pyridine at less than 15°.

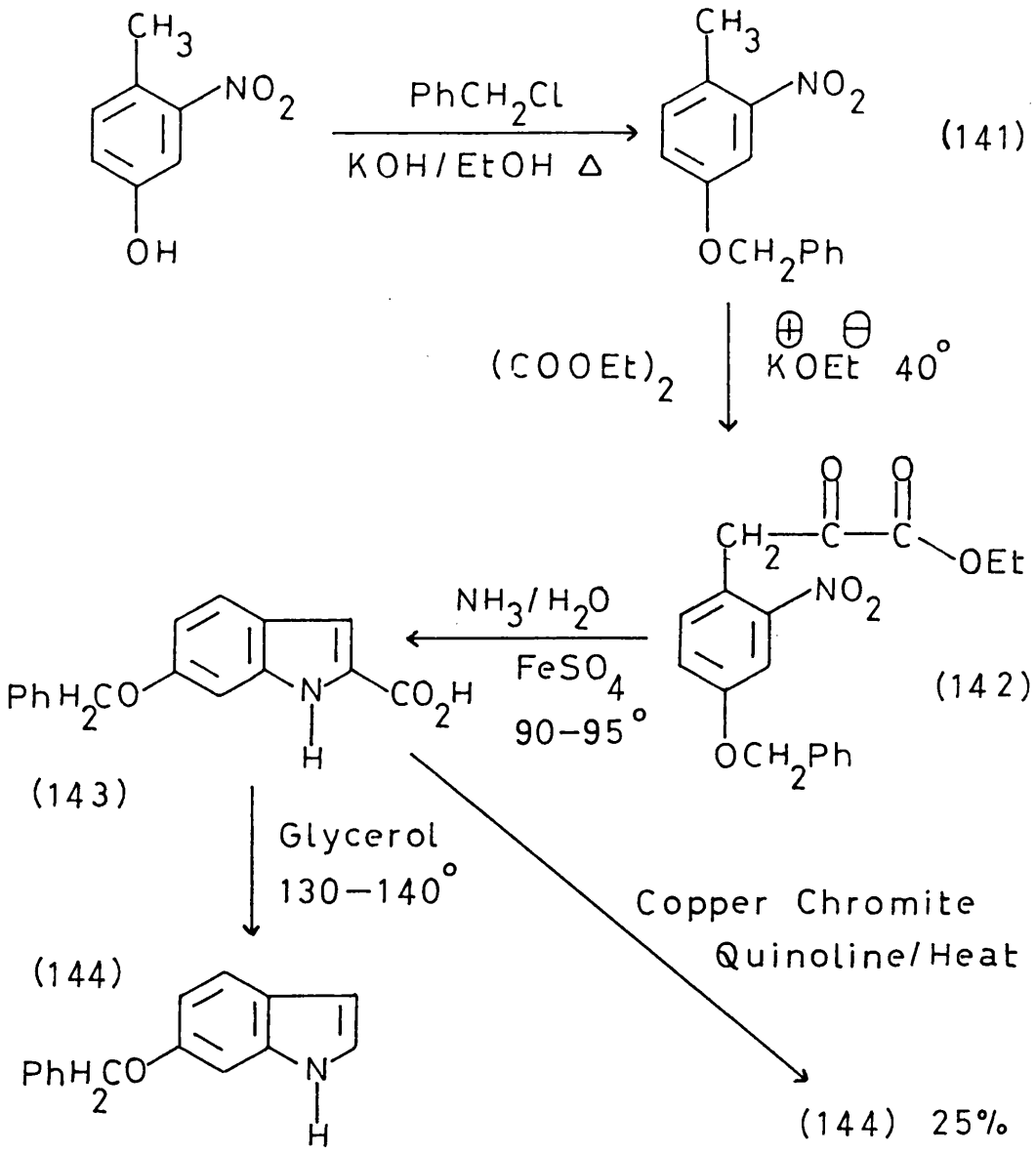


(140)

After removal of the pyridine under reduced pressure and distillation of the resulting oil in vacuo the product was obtained as a colourless, viscous liquid in 96% yield. Nitration of (140) using the method employed by Mason[94], then gave 4-methyl-3-nitrophenol in 67% yield, which is a considerable improvement with respect to the original procedure.

Having built up a stock of 4-methyl-3-nitrophenol, it was reacted with benzyl chloride in ethanolic potassium hydroxide[95] at reflux temperature to give the benzyl ether (141). This compound proved to be extremely difficult to crystallize, and the product oil required as much as five days at a temperature of -20° before crystallization commenced. Subsequently, we found that this problem could be avoided by trituration of the oil with light petroleum ether, this yielded an amorphous solid, which could then be crystallized from a 1:1 mixture of petroleum ether (b.p. 40-60°) and diethylether to give (141) as pale yellow plates. The final steps to 6-benzyloxyindole (144) are summarized in (Scheme 39).

Scheme 39



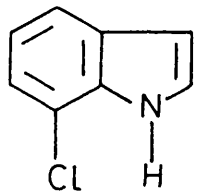
The benzyl ether (141) was condensed with diethyloxalate in the presence of potassium ethoxide to give the pyruvate ester (142). Burton and Stoves[89] reported that the acid of (142) was formed at this stage, but several repetitions of this reaction gave only the ester, in 54% yield, after purification. Once again the ester was difficult to crystallize, and trituration with diethylether followed by crystallization from ethanol was

the only rapid method of obtaining a pure product. We did not attempt hydrolysis of the ester (142) as a separate step because the next stage of the synthesis, involving reductive ring closure, employs ferrous sulphate and aqueous ammonia solution at 90-100^{o} for thirty minutes. This we felt would cause simultaneous hydrolysis, which in fact proved to be the case. However, the yield of the indole-2-carboxylic acid (143) was only 32%. A considerable amount of brown polymeric material was formed, and repeated extraction of the ferric oxide filter cake with hot dichloromethane was required.

Reducing the time taken for the reaction to twenty minutes and/or the temperature to 75-80^{o} did not improve the yield. Due to the high proportion of degraded material isolated from these reaction mixtures it seemed unlikely that the low yields were due to an incomplete reaction, however, the experiment was again repeated, but this time at the reflux temperature of 115-120^{o}. The results confirmed our expectations, as only dark polymeric material was obtained.

Despite these limitations we set about decarboxylating the indole-2-carboxylic acid (143). It was realised that we could not make use of methods involving strong mineral acids[96] because of the acid sensitivity of the benzyl phenyl group[70]. Accordingly we first attempted to decarboxylate the acid (143) by heating it with copper chromite (obtained commercially), in redistilled quinoline in a similar manner to that used successfully by

Rydon and Tweedle.[97] However, a disappointingly low yield, (25%), of the required 6-benzyloxyindole was obtained in a pure state after chromatography and recrystallization. We felt that the unsatisfactory results obtained with this method were connected with the benzyloxy group, because a similar reaction has been used successfully in this laboratory, to prepare 7-chlorindole (145).



(145)

We next turned our attention to the decarboxylation procedure favoured by Burton and Stoves[89]. The dry acid (143) was dispersed in glycerol and held at a temperature of 215-230^{o} for twenty five minutes, the solution was cooled, diluted with water and extracted with ether. This gave a red oily residue which was extracted again with hot petroleum ether (b.p. 80-100^{o}). Concentration and sublimation in vacuo gave the required indole as a white solid which was crystallized from light petroleum ether in 34% yield, but we still felt that this technique was unsatisfactory because of the extensive purification procedures required. Finally, the reaction was carried out at a temperature of only 140-160^{o} for ten minutes. This gave the product in a

sufficiently pure state to be crystallized directly, without recourse to extraction procedures, and improved the yield to 47%.

During these decarboxylation reactions, unchanged acid was always recovered upon acidification of the aqueous phase . Such findings are in accordance with those of Robertson et al[90] and Bergel and Morrison[98] who also failed to achieve a complete reaction.

Robinson and co-workers[99] have reported that fusion of the ammonium salt of 6-methoxyindole-2-carboxylic acid is a high yielding method for the preparation of 6-methoxyindole. However, we rejected the application of this method of decarboxylation to our synthesis of 6-benzyloxyindole because this compound is reported to be temperature sensitive[85], and there is a lack of documentation for this approach to benzyloxyindoles in the literature.

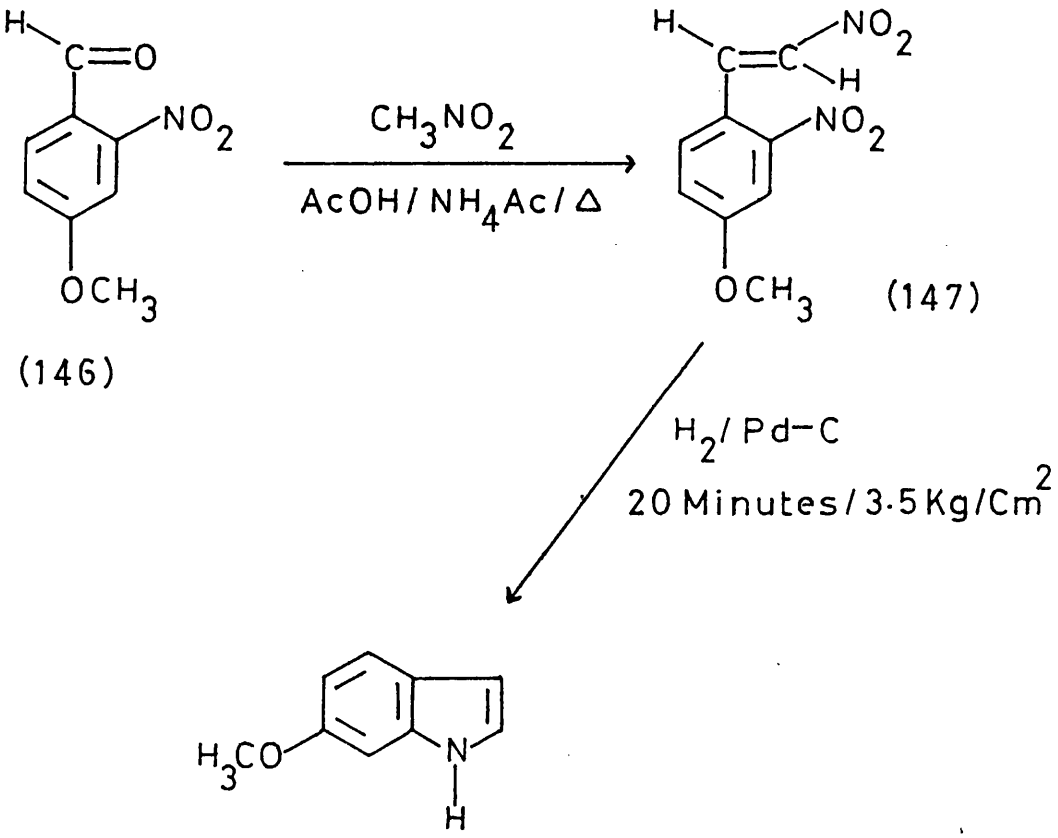
At this stage we regretfully decided that due to the difficulties discussed above, and the low yields obtained, it was unprofitable to continue in our attempts to synthesize (10-20 g) quantities of 6-benzyloxyindole by this method.

We now turned our attention to the synthesis of 6-methoxyindole, this compound is an equally good substrate for our proposed Grignard condensation (see p. 76), and would, we hoped, overcome

some of the difficulties previously discussed. It was realised that in this case it would not, of course, be so easy to de-alkylate the substituent at a later stage of the synthesis. However, the literature contains a number of efficient modern methods[62,100,101] that can be applied to solve this problem.

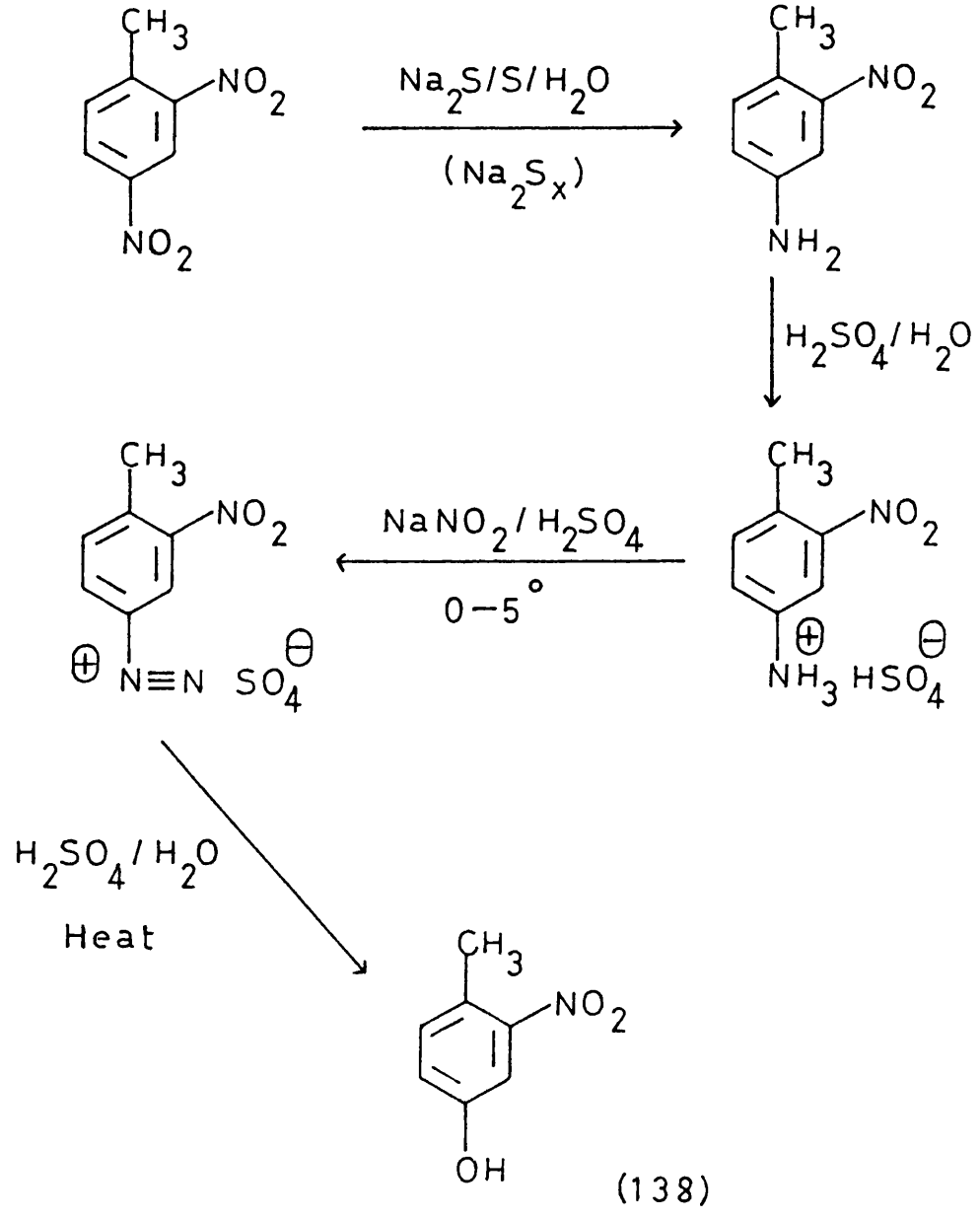
Having made the decision to abandon the benzyloxy group as a protecting function and to use the methoxyl unit instead, a much more efficient method of synthesizing the required indole was now open to us via the appropriate 2,β-dinitrostyrene (147), which on catalytic hydrogenation should ring close to the corresponding indole,[102,103] as indicated in (Scheme 40).

Scheme 40

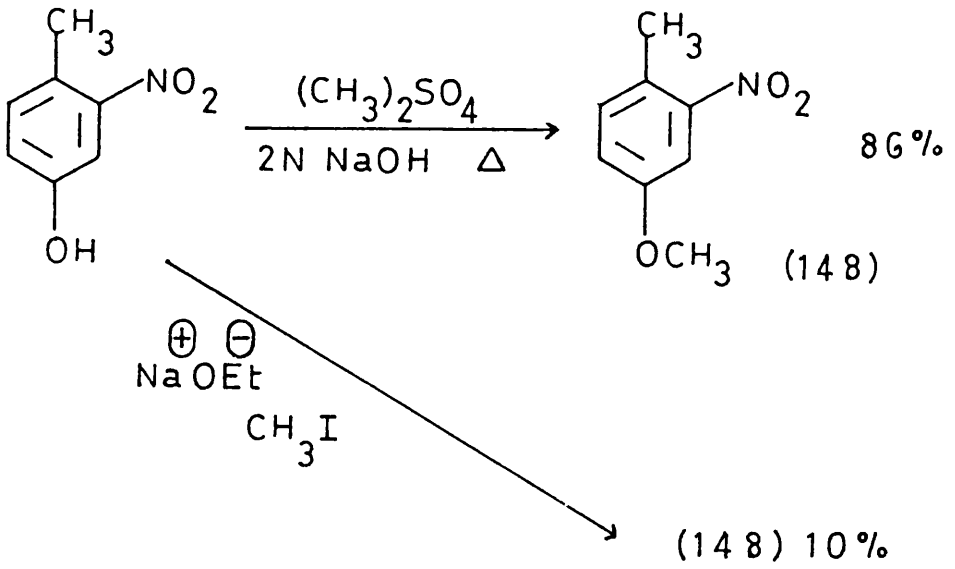


This route required the preparation of fairly large (30-40g) quantities of the aldehyde (146). This compound was not commercially available so a route to it was sought. We decided to use 4-methyl-3-nitrophenol as the starting material, but it was desired to improve the preparation of this, with respect to that previously employed (see p.. 92). The work that followed is outlined in (Scheme 41).

Scheme 41.



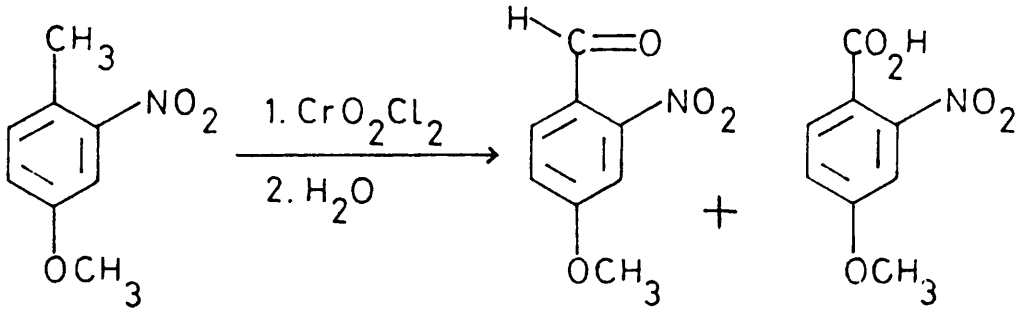
(138)



We began with the inexpensive, commercially available 2,4-dinitrotoluene. The 4-nitro group was selectively reduced using sodium polysulphide[104] and the resulting amine diazotized *via* the sulphuric acid salt[105]. Hydrolysis in boiling aqueous acid then gave the desired 4-methyl-3-nitrophenol in 63% yield. A great improvement compared with previous attempts. This phenol was then methylated with dimethylsulphate in the presence of 2N sodium hydroxide solution,[106] after extraction with chloroform and distillation under reduced pressure the pure methyl ether (148) was obtained as a pale yellow mobile liquid in 84% yield. Attempts were made to carry out this reaction using methyl iodide as the methylating agent, but this reagent gave only a 10% yield of the desired ether (148).

Having obtained a good stock of the compound (148) various methods were attempted to convert it to the desired aldehyde.

We first tried Et'ards oxidation with chromyl chloride,[107] but this gave a considerable amount of the acid (149) due to over oxidation.



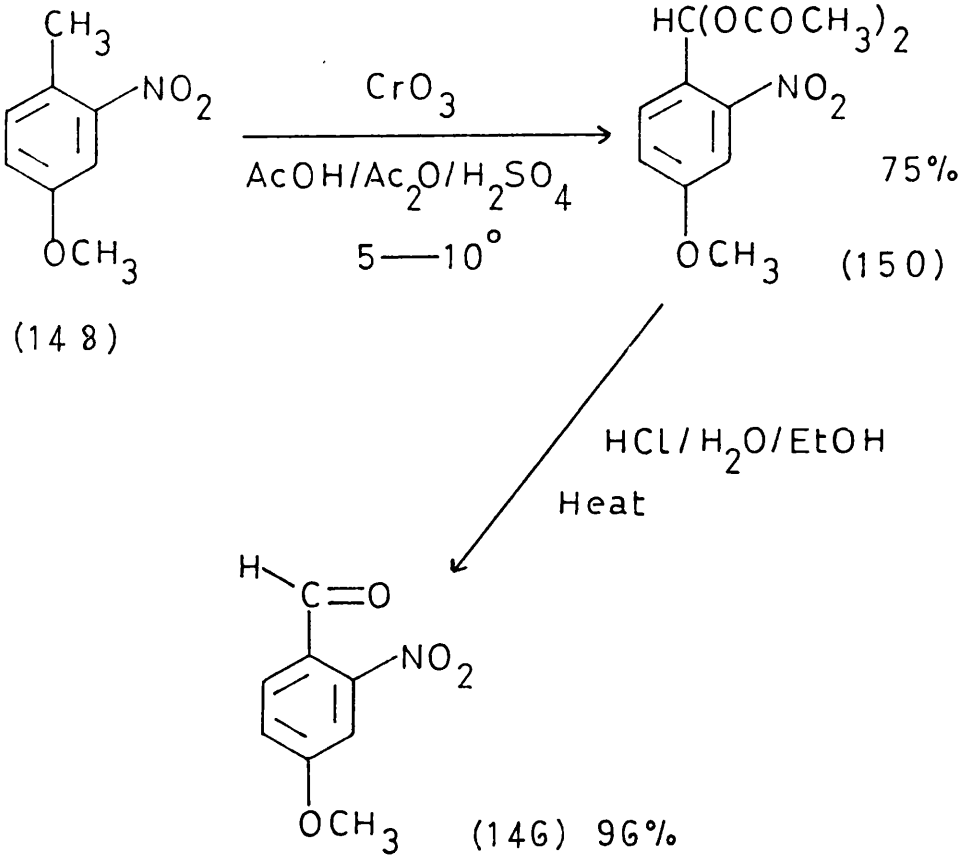
(146) (149)

Minor product Major product

Attempts were made to overcome this problem by rapid extraction or steam distillation to remove the desired aldehyde before it could oxidize, but this failed to improve the yield of (146).

A better method proved to be the use of chromic trioxide in a mixture of acetic anhydride and acetic acid containing a small amount of sulphuric acid at 5-10°.[108] The product of this reaction is the gem-diacetate (150), thus over oxidation is inhibited. The compound (150) was isolated and subsequently hydrolysed with aqueous ethanolic hydrochloric acid to give an overall yield of 70% based on the methyl ether (148), as outlined in (Scheme 42).

Scheme 42.



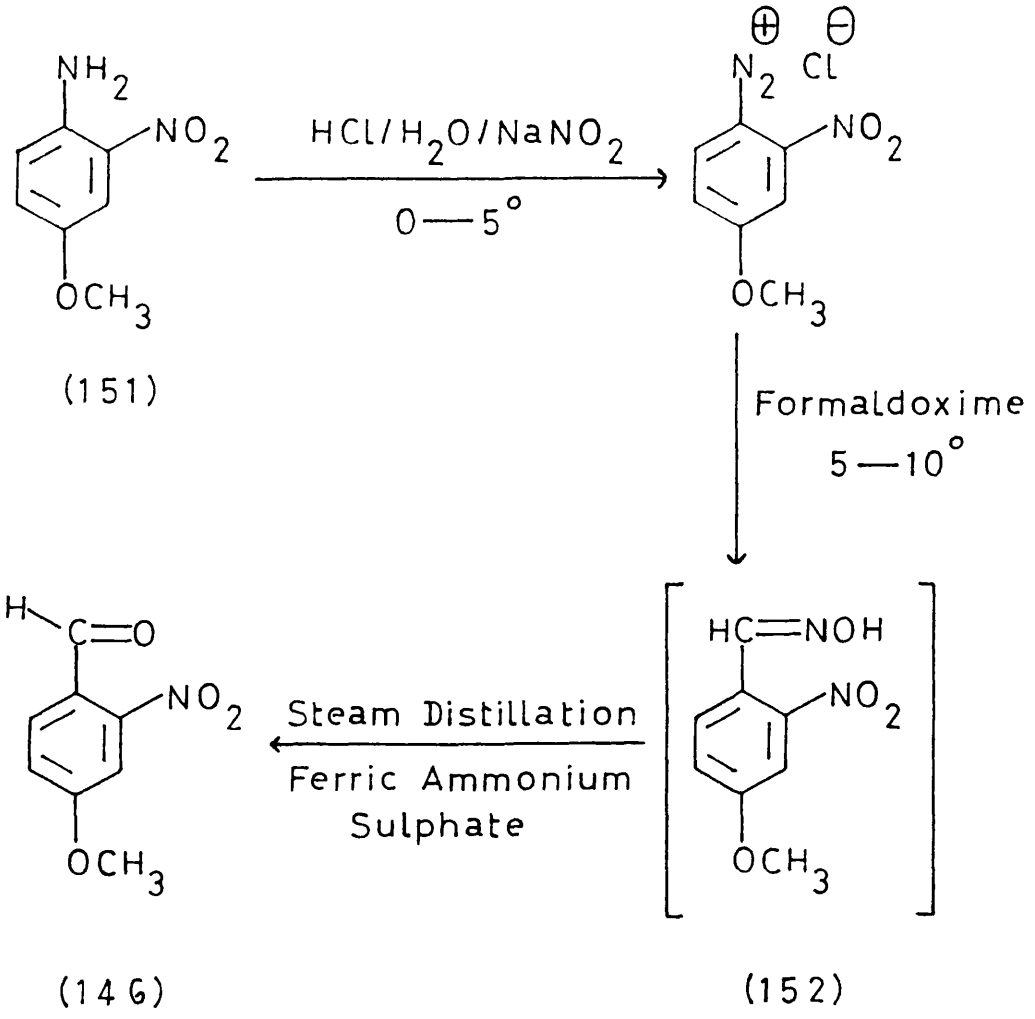
A stock of the aldehyde (146) was prepared and converted to the 2,β-dinitrostyrene (147) by heating in glacial acetic acid with nitromethane in the presence of sodium acetate[109]. This base condensation system gave a 74% yield of the desired product, but the more traditional method employing aqueous sodium hydroxide as the base[110], failed to condense this para-methoxyaldehyde, as did potassium ethoxide.

Low pressure hydrogenation[109] of the 2,β-dinitrostyrene (147) over palladium/carbon in a mixture of acetic acid and ethylacetate proceeded smoothly to give a 85% yield of pure 6-methoxyindole after crystallization.

Reductive ring closure of the 2,β-dinitrostyrene (147) with iron and acetic acid[78] at 90-100^{o} was also attempted, in an effort to overcome the problems posed by our limited capacity hydrogenator when dealing with a compound such as this, which exhibits a limited solubility in the solvent system of choice. However, in line with our previous expectations (p. 85.), this method proved to be much less satisfactory than catalytic hydrogenation, because the product required treatment with charcoal, followed by chromatography and crystallisation before it reached a sufficiently pure state to be used in the next stage of the synthesis.

The overall efficiency of this indole systhesis, was still further improved at a later stage by employing Woodwards[109] excellent, if somewhat time consuming method of preparing the aldehyde (146). This approach uses the commercially available amino compound (151) as the starting material. Treatment of this with an ice-cold solution of formaldoxime gives the intermediate 2-nitroanisaldoxime (152); this is hydrolyzed by steam distillation in the presence of a large quantity of ferric ammonium sulphate. The desired aldehyde (146) is obtained by filtration of the distillate. This reaction sequence is outlined in (Scheme 43).

Scheme 43



(151)

(146)

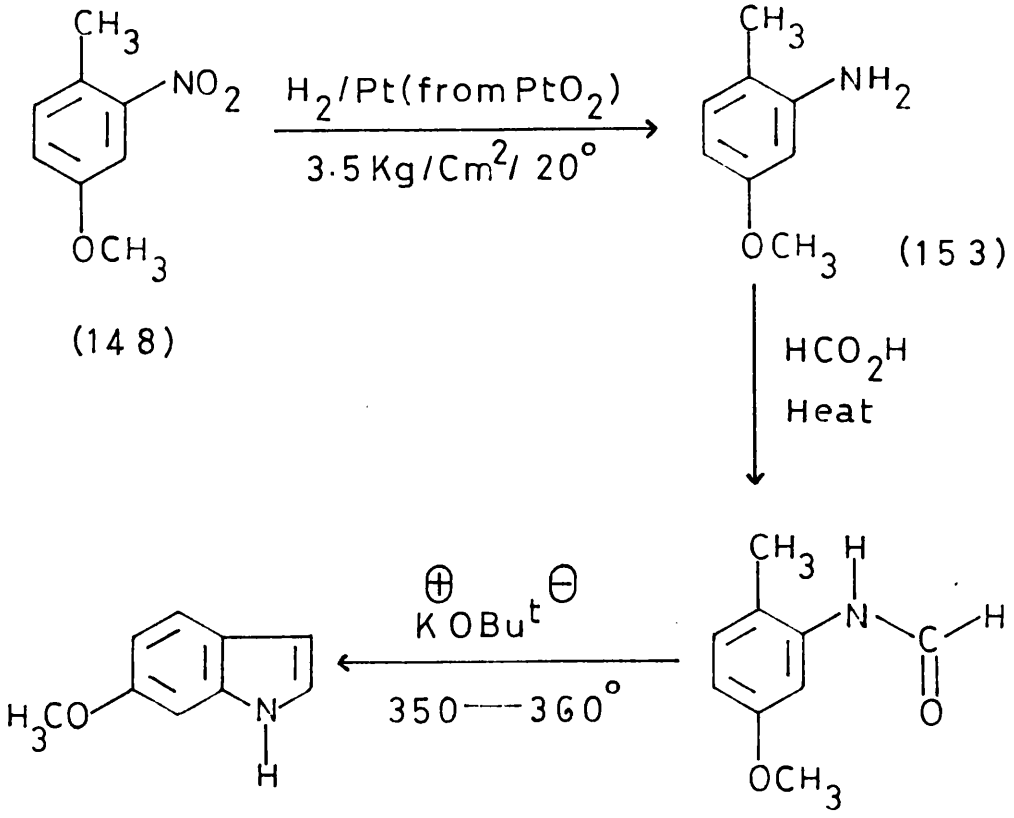
(152)

Working on a (120g) scale the steam distillation required sixteen hours to obtain the maximum yield of (146) (81g, 55%). The first fractions were treated with sodium bisulphite solution in order to separate the aldehyde from a considerable amount of 3-nitroanisole. Despite these minor difficulties, the method proved to be the best for producing (+10g) quantities of 6-methoxyindole.

Before leaving the topic of 6-methoxyindole synthesis it is noteworthy to mention two other somewhat more speculative attempts that were made to prepare this compound.

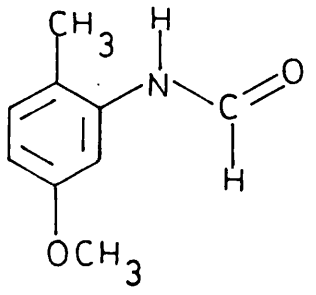
Firstly, we attempted a Madelung synthesis as outlined in (Scheme 44), despite our reservations concerning the presence of the deactivating para-methoxy substituent (see p. 84). This we did because the starting material (148) was already available and there are reports of successful Madelung syntheses in the literature giving acceptable yields of product, for example, 2-ethyl[111], 5-methyl[112] and many other alkyl indoles have been prepared in this way. The yields range from 30-75%, although it should be said that this is with alkyl, rather than alkoxyl groups present.

Scheme 44.

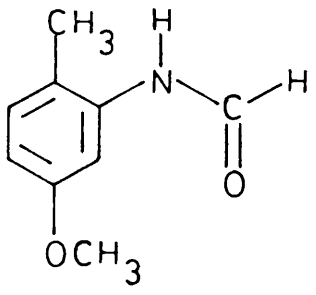


4-Methyl-3-nitroanisole was reduced to 6-methyl-3-anisidine (153) by low pressure hydrogenation using Adams platinum oxide catalyst. This gave a 96% yield of (153) as shining amber coloured cubes from ethanol. Condensation of this compound with 98-100% formic acid[74], gave a 84% yield of the formamide (154) as a white microcrystalline solid.

When the 100 MHz ^{1}H n.m.r. spectrum of this compound was determined, two sets of clearly defined, and closely spaced signals were observed for each resonance, the spectrum is reproduced in (Figure 1). The formamide (154) clearly exists as a mixture of the E- and Z-forms shown, at room temperature, due to restricted rotation about the amide N–C bond.



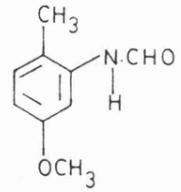
E



Z

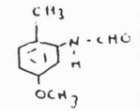
The chloroform spectrum at room temperature shows the two sets of methyl and methoxyl protons in a ratio of approximately 57:43, and the two N–H signals are separated by 0.4 p.p.m., the more abundant of which occurs at lowest field. From this we concluded that the Z-form is favoured at room temperature, as the N–H signal of the E-form is shifted to higher field as it will experience the anisoptropic shielding effect of the formyl carbonyl function.

(Figure 1)



(E)-and (Z)-forms

SPECTRUM NO. 3468
DATE 3.1.76
FREQ. 100 MH
NUCLEUS H
SAMPLE Del Nmf



SOLVENT CDCl3
CONC. 32 mg
REFERENCE TMS
LOCK FX7
TEMP 20 °C
R.F. LEVEL 46
A.F. LEVEL ANALYTICAL 6 6
LOCK
SD
AMPLITUDE
ANALYTICAL X10 4
LOCK
INTEGRATOR
FILTER 16
OFFSET Hz PPM
—FREQ.—FIELD/FREQ.—FIELD
OPERATOR [illegible]
REMARKS
No peaks > 10 ppm

SWEEP TIME (SEC.)

25	50	100	250	500
1000	2500	5000	10000	

SWEEP WIDTH (Hz) (X0.01PPM)

27	54	108	270	540
1080	2700	5400	10800	

WIDE SWEEP (GAUSS)

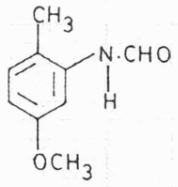
10.8	27	54	108	540

Printed in U.K.
Chart No. 4H-C

Supplied by Nuclear Magnetic Resonance Ltd., Magnetic House, Scrubbs Lane, Bledlow Ridge, High Wycombe, Bucks.

JEOL

(Figure 2)



Energy barrier to free rotation exceeded

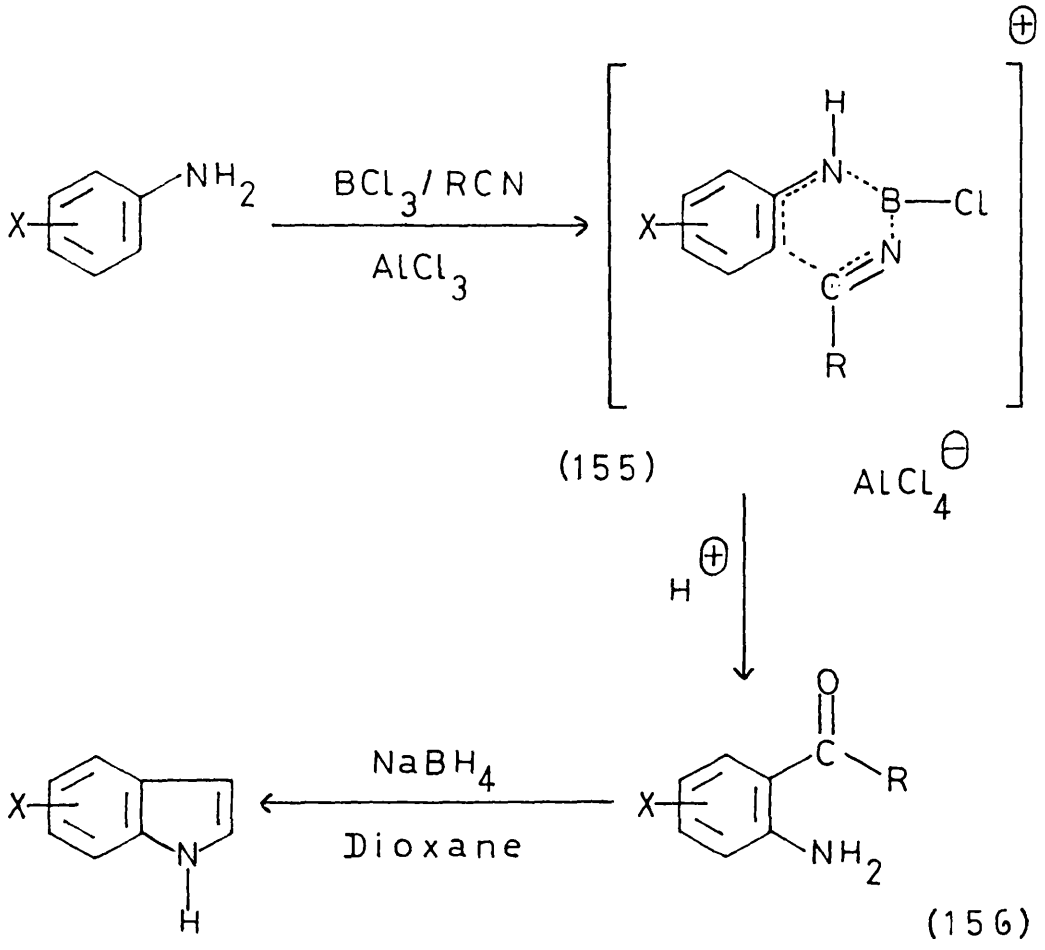
| | | |

To gain further evidence in support of this, we carried out a variable temperature n.m.r. experiment. This required a higher boiling solvent than chloroform so d_6 dimethylsulphoxide was chosen. Firstly, the spectrum was run at room temperature in this solvent in order to take account of any changes due to its greater polarity. The spectrum was then repeated at 100°, and clearly shows (Figure 2), that the signals have coalesced to single resonances when the energy barrier to free rotation about the amide bond was exceeded.

When the formamide (154) was heated with potassium tertiary butoxide at 350-360° for twenty minutes a dark brown solid was obtained. This was purified by treatment with charcoal, followed by column chromatography on basic alumina, eluting with ether. The eluted product was then sufficiently pure to be crystallized directly from light petroleum ether. However, the yield of 6-methoxyindole was only 10%. The poor yield of this reaction and the high proportion of accompanying breakdown products, (starting material was never isolated), lead us to attempt to optimize the conditions by reducing their severity. However, decreasing the reaction temperature to 180-200° did not improve the yield, and this approach was abandoned.

At this time an interesting paper by Sugasawa and his collaborators was published[113]. These workers prepared various substituted indoles by the method outlined in (Scheme 45).

Scheme 45.



Where: (R = CH_2-Cl, or alkyl or aryl when this method is used to synthesize 2-aminoketones, rather than indoles), and (X = H, alkyl, halogen or O-alkyl, although the method is probably not restricted to these functions).

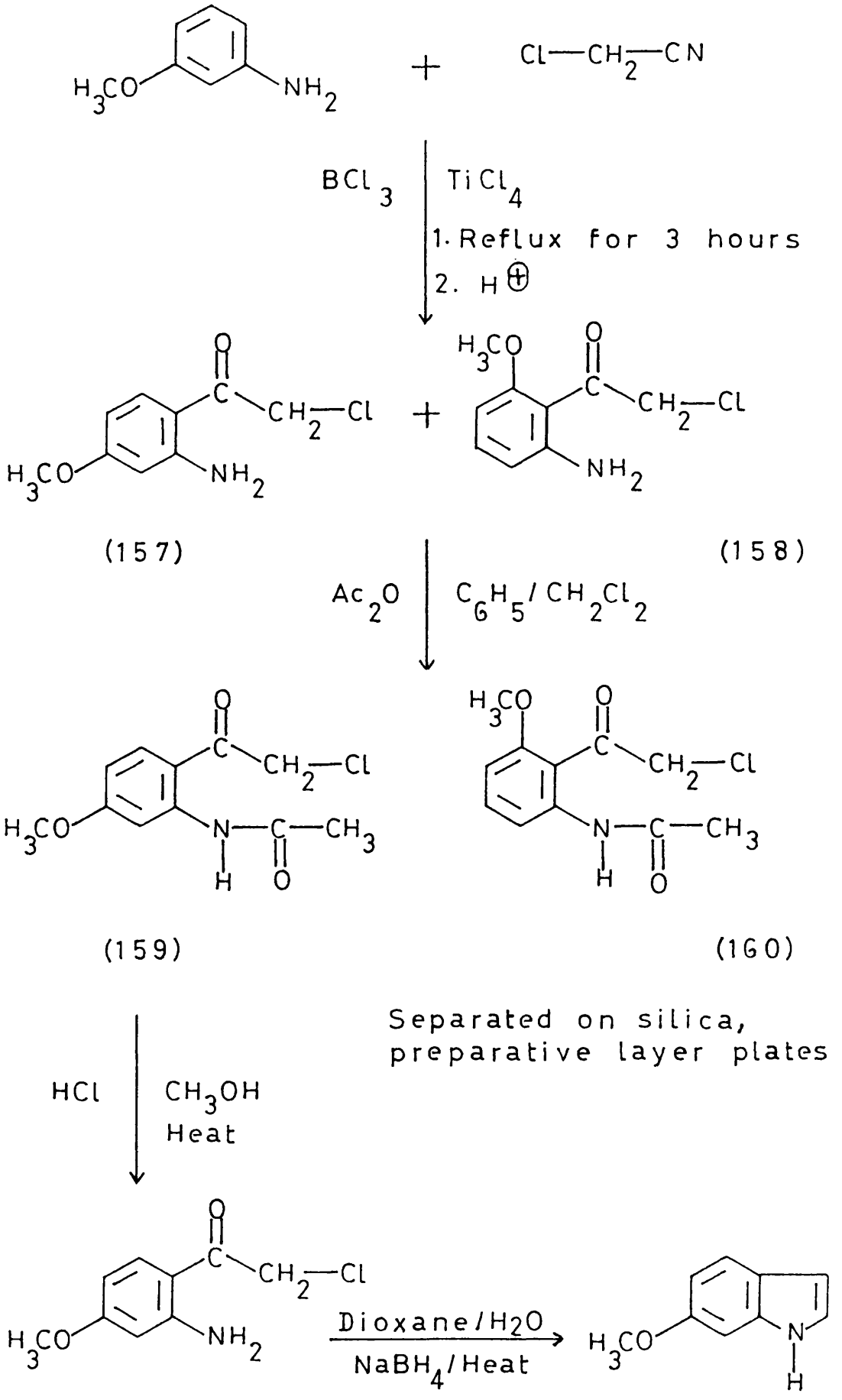
When this route is used to synthesize indoles the appropriate aniline is condensed with chloroacetonitrile, in the presence of boron trichloride and aluminium trichloride. The reaction is thought to take place *via* a cyclic transition state involving a boronium cationic species of type (155) stabilized by the tetrachloroaluminate anion. Treatment with acid then furnishes the corresponding 2-amino-α-chloroacetophenone (156), and

reductive cyclization with sodium boronhydride in dioxane completes the sequence to the desired indole. The synthetic versatility of this route offers a wide range of substituted indoles and has been used in this laboratory to prepare 7-chloroindole in 70% overall yield. However, in this case a 2-substituted aniline is the starting material and the structure of the product is unambiguous.

Unfortunately in the case under study two isomers, both the 4- and 6-methoxyindoles may result, but, as both of these compounds would be useful to us we attempted this reaction beginning with 3-anisidine. We also replaced aluminium trichloride with titanium tetrachloride to minimize the possibility of the methoxyl function undergoing cleavage. The work that followed is outlined in (Scheme 46).

The reagents were mixed together in dry benzene under a vacuum at 0-5^{o}, and then heated at reflux temperature for three hours. After cooling to 0^{o}, the boronium cation was opened by treatment with ice-cold, dilute hydrochloric acid and the resulting ketamine hydrolysed by heating the mixture at 80^{o} for thirty minutes. When cool, the pH of the solution was adjusted to 3-4. Extraction with dichloromethane then gave the expected mixture of 4- and 6-methoxy-2-amino-α-chloroacetophenones (157) and (158).

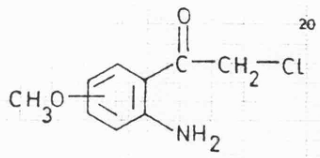
Scheme 46.


Cl—CH2—CN
BCl3
TiCl4
1. Reflux for 3 hours
2. H⊕
(157)
(158)
Ac2O
C6H5/CH2Cl2
(159)
(160)
Separated on silica, preparative layer plates
HCl
CH3OH
Heat
Dioxane/H2O
NaBH4/Heat

Comparison of the integrals for the methylene groups in the ^{1}H n.m.r. spectrum of the mixture (Figure 3), suggested a product ratio close to 1:1, which was confirmed by gas chromatography. T.L.C. on basic alumina plates, eluting with a mixture of benzene and light petroleum ether in a ratio of 7:3 showed a slight difference in Rf values, as did neutral alumina or silica plates with the same solvent system. Various other solvent systems were used (see experimental section, p.274), but none of them improved the separation. Column chromatography was attempted using basic alumina and the same solvent system as before, but an efficient separation could not be realized despite much effort. Basic alumina preparative plates were prepared and once more eluted with a mixture of benzene and light petroleum ether, but this time in a ratio of 3:2. However, this also failed to separate the mixture, and we noted that Sugasawa had experienced this problem. Sugasawa reports a small scale separation of these isomers using a mixture of n-hexane and benzene in a ratio of 2:1, but he does not quote which support material was used and we were unable to obtain an efficient separation in this way with any of the chromatographic supports available in our laboratory.

Reduction of the mixture, without separation gave the expected 6- and 4-methoxyindoles in high yield ($\sim$ 85%) and a product ratio of 2:1 in favour of the desired isomer. Unfortunately these isomers proved to be almost identical in chromatographic properties and could not be separated satisfactorily.

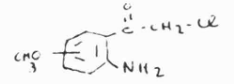
(Figure 3)



~ 1:1 Mixture of o-and p-methoxy isomers

DEUTERATED

SPECTRUM NO. DA124
DATE 22-6-79
FREQ. 100 MHz
NUCLEUS H
SAMPLE



o and p 1:1 MIXTURE

SOLVENT	CDCl3
CONC.	22 MG
REFERENCE	TMS
LOCK	EXT
TEMP.	32 °C
R.F. LEVEL	46
A.F. LEVEL ANALYTICAL	4-2
LOCK	
SD	
AMPLITUDE	
ANALYTICAL	X16-6
LOCK	
INTEGRATOR	
FILTER	16 Hz
OFFSET	PPM

FREQ. FIELD/FREQ. FIELD

OPERATOR Dickwood

REMARKS

a, b, c, are 3 repeat integrals of the $-CH_2-$ groups

SWEEP TIME (SEC.)

25	50	100	250	500
1000	2500	5000	10000	

SWEEP WIDTH (Hz) (X0.01PPM)

27	54	108	270	540
1080	2700	5400	10800	

WIDE SWEEP (GAUSS)

10.8	27	54	108	540

Printed in U.K.
Chart No. 4H-C

Supplied by Nuclear Magnetic Resonance Ltd., Magnetic House, Scrubbs Lane, Bledlow Ridge, High Wycombe, Bucks.

JEOL

We next N-acetylated the mixture of (157) and (158) and found that the resulting N-acetyl compounds (159)and (160) could be more easily separated on silica gel preparative plates, eluting with dichloromethane. This technique gave pure 4-methoxy-2-acetamido-α-chloroacetophenone in a 79% recovery at a loading of 100 mg per plate.

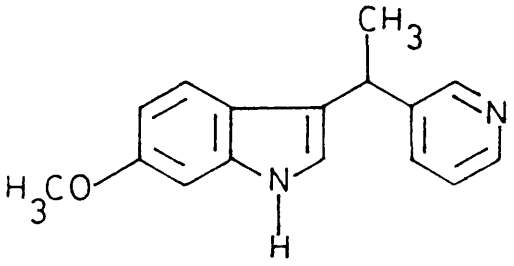
Hydrolysis in dry methanol saturated with hydrogen chloride gas furnished pure 4-methoxy-2-amino-α-chloroacetophenone in excellent yield ($\sim$ 92%).

When reduced with sodium borohydride in aqueous dioxane at elevated temperatures this compound gave 6-methoxyindole in a smooth, clean, high yielding reaction ($\sim$ 84%). Unfortunately the quantities of this indole required (10-15g) would necessitate the preparation and expenditure of 50-60 preparative T.L.C. plates, which is clearly unprofitable.

It was concluded that Sugasawa's method is an excellent two stage indole synthesis of value when the substituent in the aryl ring is located ortho or para to the amino function so that isomer problems are eliminated. It may be possible in other cases, with different substituents to leave the separation stage until after indole formation has been effected, but as stated above, in the case of the methoxyindoles this is not satisfactory.

By this time however, we had developed the more productive procedure summarized in (Scheme, 40 p. 97), and built up a stock of 6-methoxyindole. As one may expect this indole proved to be far more sensitive to oxidation and acid catalysed polymerisation than indole itself[86], due to the increased electron availability provided by the 6-methoxy substituent. This property necessitated rather more careful handling than is normally required with indoles. The compound was stored in the dark, under nitrogen at -18^{o}, and under these conditions no decomposition occurs, even after several weeks. On the other hand a sample exposed to the atmosphere and to light at room temperature was seen to darken in a matter of days. Any attempt to react this indole in the presence of mineral acids results in the rapid formation of dark-red, intractable, polymeric material.

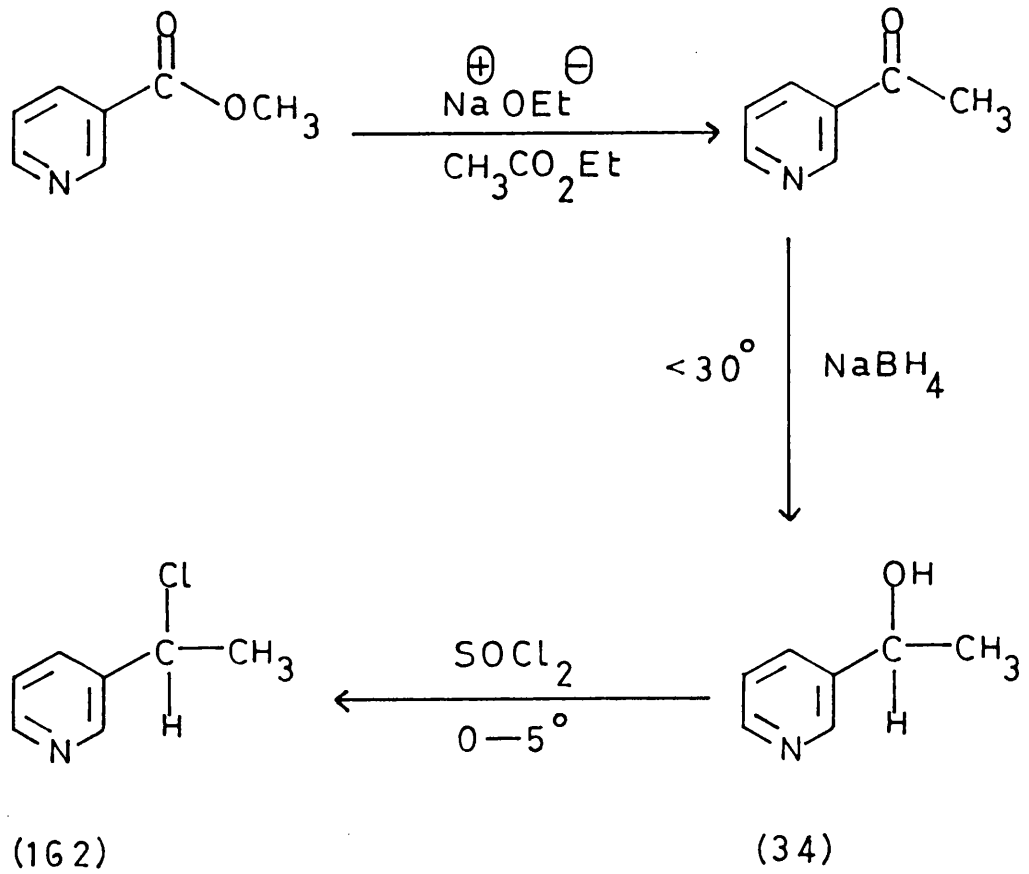
We now turned our attention to the problems concerned with the formation of the indolylmagnesium bromide of this indole and its condensation with 3-(1-chloroethyl)pyridine in a Grignard reaction, to give 3-[1-(3-pyridyl)ethyl]-6-methoxyindole (161).



(161)

Firstly, 3-(1-chloroethyl)pyridine was prepared by the series of reactions shown in (Scheme 47).

Scheme 47.



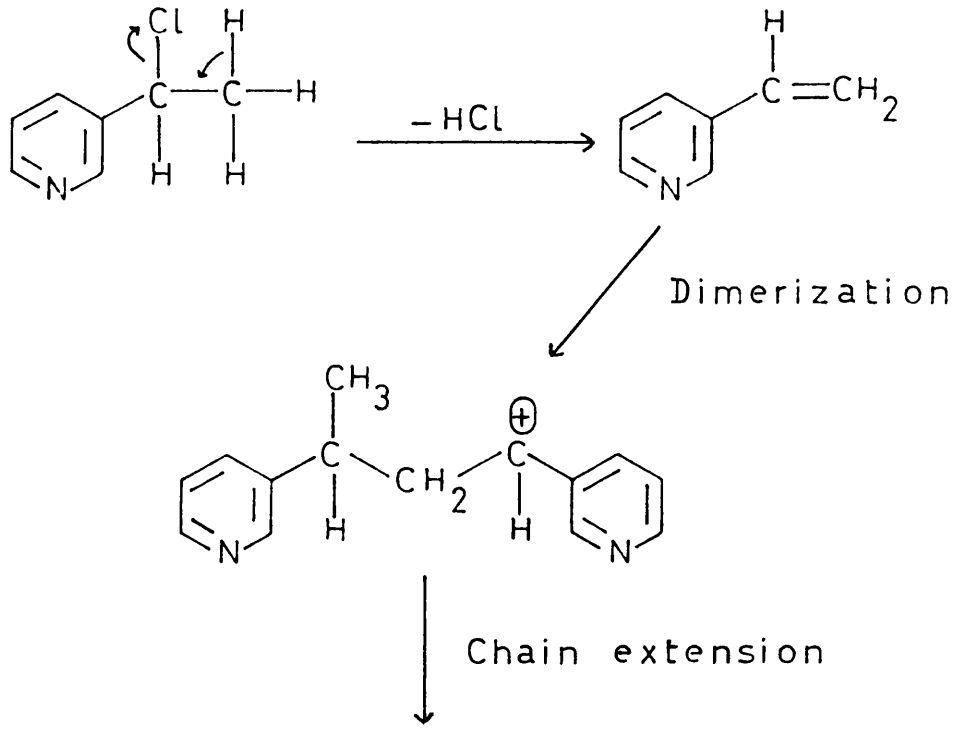
3-Acetylpyridine is a quite expensive compound, so with an eye to economy we prepared it from the much cheaper methylnicotinate using a modification of the method employed by Kolloff and Hunter[114]. Here, the ester was condensed with ethyl acetate in the presence of sodium ethoxide and the condensation product hydrolysed and decarboxylated with hot aqueous hydrochloric acid. After

basification, extraction and distillation under reduced pressure, 3-acetylpyridine was obtained in 76% yield. Reduction of 3-acetylpyridine with sodium borohydride in ethanol at less than 30^{o} gave 3-(1-hydroxyethyl)pyridine (34) as a colourless, viscous liquid in 98% yield after distillation under reduced pressure. The alcohol (34) was then reacted with thionyl chloride at less than 10^{o}, followed by basification and extraction at $0 - 5^{o}$. This treatment gave 3-(1-chloroethyl)pyridine (162) as an unstable yellow liquid in 96% yield.

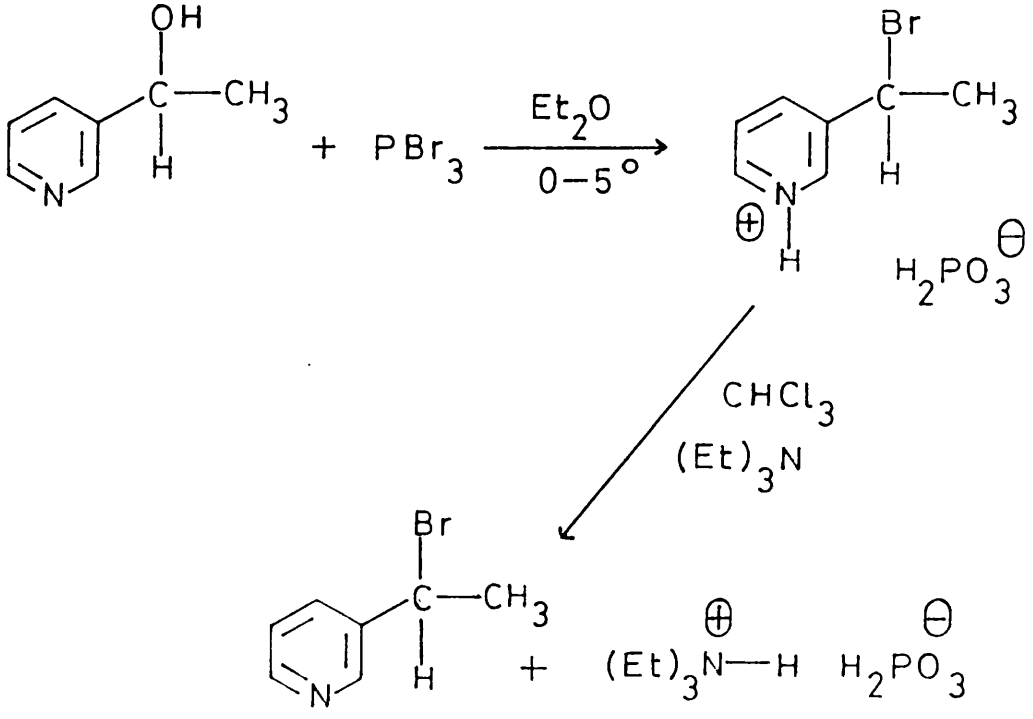
Previous synthetic work aimed at producing ellipticine precursors via the Grignard reaction between various indolyl magnesium bromides and 3-(1-chloroethyl)pyridine to give indolylpyridylethanes of the general type (125) (see, p. 76), has proved most frustrating. A yield no greater than 50% was ever achieved and with some indole substituents as little as 25-30% of the desired product was obtained. This has previously been rationalized, as being due to elimination reactions of 3-(1-chloroethyl)pyridine leading to 3-vinylpyridine which may dimerize and form higher polymers as outlined in (Scheme 48).

Confirmation of the facile nature of this type of reaction has been obtained in this laboratory by the author. In a bid to increase the reactivity of the haloethyl pyridine we prepared 3-(1-bromoethyl)pyridine from the hydroxy analogue as shown in (Scheme 49).

Scheme 48



Scheme 49

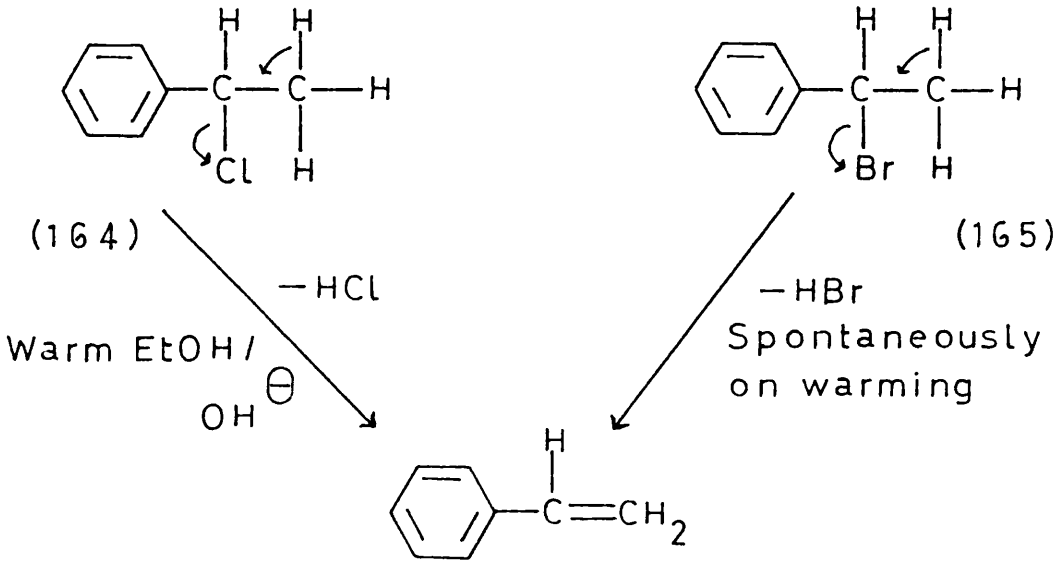


(163)

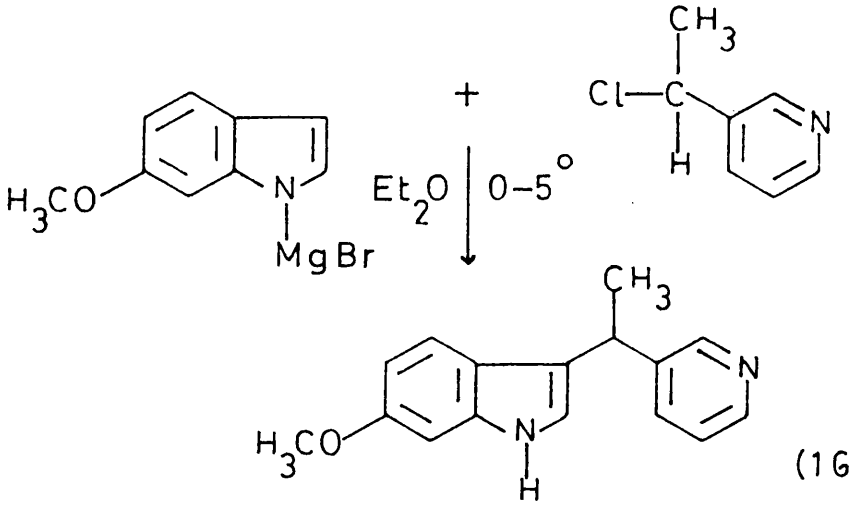
The bromo compound (163) proved to be even more unstable than its chloro analogue. On standing at room temperature for a few hours the viscosity increased rapidly and the colour changed from light yellow to dark orange Investigation of this residue showed it to be mainly a mixture of 3-vinylpyridine, dimers and higher polymers. This may be compared with the chloro analogue which will survive up to two days at room temperature. In view of this sensitivity the bromo compound (163) was stored at -20^{o}, but even at this temperature decomposition occurred within forty eight hours, compared with approximately two months for the chlorinated compound.

In view of the fact that the bromide ion is generally considered to be a better leaving group than chloride these results are to be expected, and eliminations of this type are well known in the benzene series. Hence 1-phenyl-1-chloroethane (164), on treatment with warm alcoholic base, readily forms styrene[115], and the bromo analogue (165), eliminates hydrogen bromide spontaneously[116], as shown in (Scheme 50).

Scheme 50



Despite our concern over the instability of 3-(1-chloroethyl) pyridine we viewed the following reaction with optimism.



(161)

We felt that 6-methoxyindole should be a more activated substrate than indole itself, and the reaction between its indolylmagnesium bromide and the chloroethylpyridine might now be more competitive than the base catalysed decomposition of the latter.

Accordingly the reaction was carried out under the usual Grignard conditions for this type of reaction,[19,54] employing a 1:1 molar ratio of the two reactants in dry ether. Due precautions for the instability of the chloroethylpyridine were made, thus the reaction was held at a temperature below 5° for the first four hours. After the usual work up procedure (see; experimental section, p. 281), the chloroform extract gave the expected indolylpyridylethane (161), but after trituration with diethylether and crystallization from ethanol the yield was only a modest 15%. However, a high proportion of 6-methoxyindole could be recovered from the ether extracts, so various attempts were made to improve the yield of the reaction.

Firstly, we attempted to optimize the reaction conditions by altering the time taken for the reaction and the temperature employed. The experiment was repeated twice, firstly at 0^{o} for the first hour followed by stirring at room temperature for a further twenty four hours, and secondly at $10\text{-}15^{o}$ but working up after only one hour. However, the products in each case were the same as those obtained in the first experiment. Due to the temperature sensitivity of 3-(1-chloroethyl)pyridine it was thought unlikely that an experiment at elevated temperatures would be of any value, but in order to complete our examination of this reaction we carried out such an experiment. The reaction mixture was heated at its reflux temperature in ether for two hours, but our fears were confirmed when it was found that a considerable amount of polymeric material together with some unreacted 6-methoxyindole resulted.

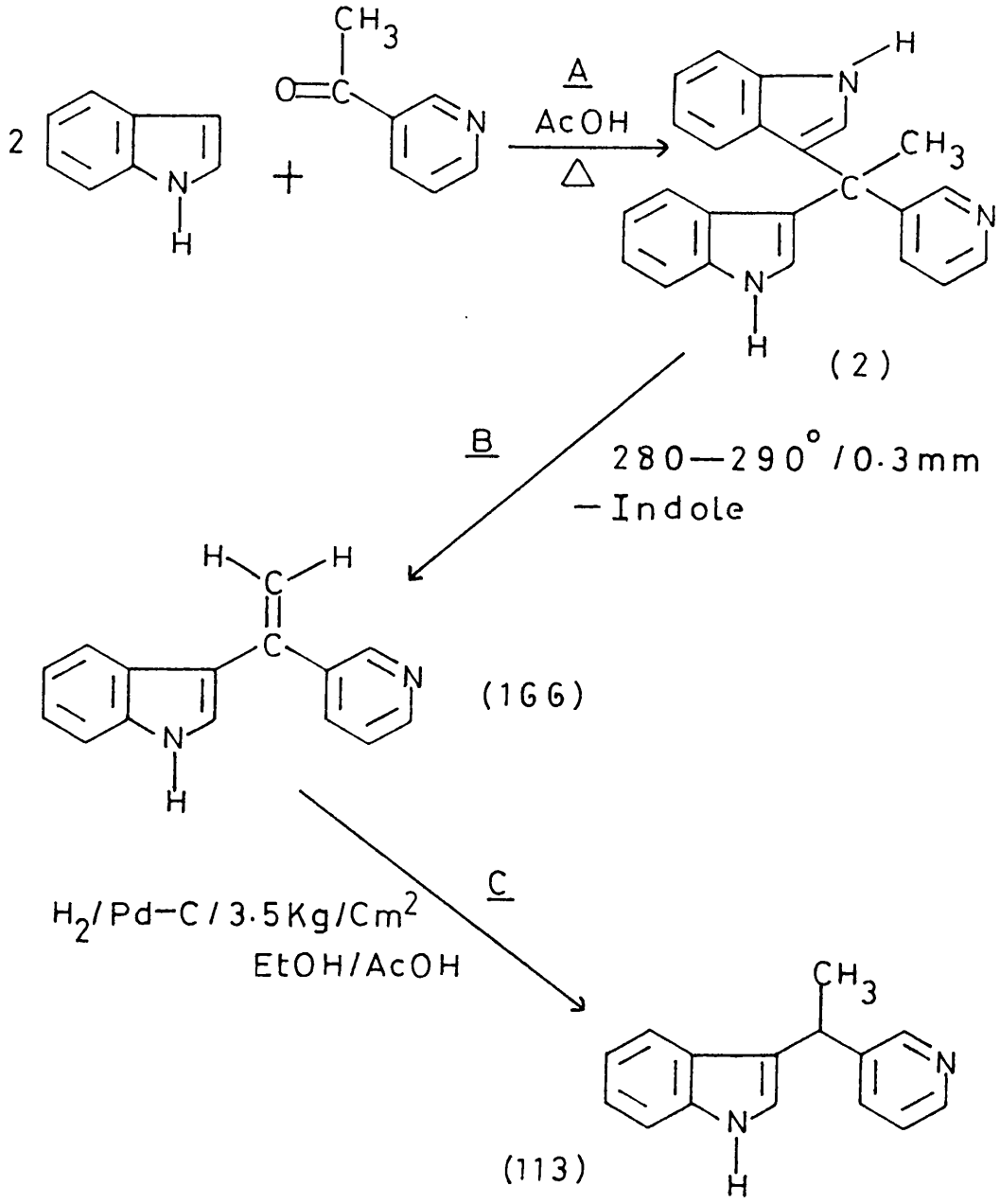
We next reverted to the more favourable conditions of the original experiment, but performed an inverse addition. The experiment is usually carried out by adding the chloroethylpyridine to an ice-cold solution of the appropriate indolylmagnesium bromide. This was reversed, in the hope of reducing the tendency for any excess alkyl halide to quaternise the pyridine nucleus, and also to ensure that the haloethylpyridine was not exposed to an excess of ethylmagnesium bromide, which might result in side reactions involving alkylation of the pyridine nucleus. However, despite these precautions no improvement in the yield of (161) could be achieved.

At this time we were unable to give a precise rationalization of the unexpectedly low yields obtained with this reaction when 6-methoxyindole is used as the substrate (see p, 178 for further work on this topic). Clearly the facile elimination of hydrogen chloride from 3-(1-chloroethyl)pyridine, previously discussed, is a major disadvantage in this reaction. Further support for the instability of this compound has been obtained by Iddles, Lang and Gregg[117], who have prepared 3-vinylpyridine from 3-(1-chloroethyl) pyridine by treating it with a strong base. These authors also comment that 3-vinylpyridine will polymerise in a period of a few hours at room temperature and very rapidly upon heating.

Disappointed by the low productivity of this approach we next embarked upon a lengthy series of experiments aimed at finding an improved method of synthesizing the required indolyl-pyridylethane (161). Due to the large amount of labour, time and expense required to prepare (10-20g) quantities of 6-methoxyindole, we decided to employ indole itself in these investigations.

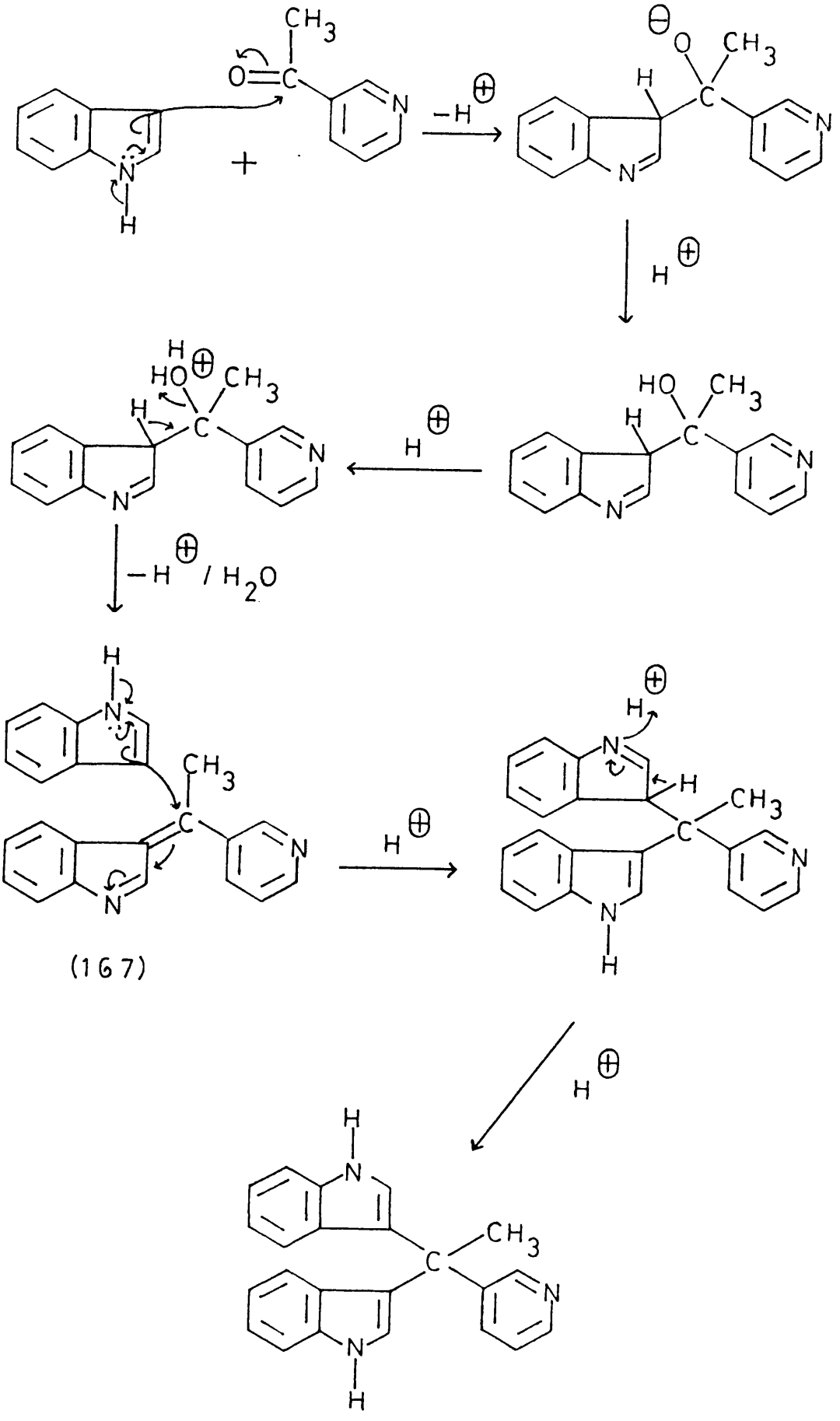
Bergman and Carlsson[6] have condensed indole and 3-acetylpyridine in a 2:1 ratio to give 1,1-bis(3-indolyl)-1-(3-pyridyl)ethane (2). Vacuum pyrolysis of compounds of this type[4], causes elimination of one of the indole groups, and an unpublished communication from Professor Bergman[118] indicated that this technique could be profitably applied to (2) which would provide 1-(3-indolyl)-1-(3-pyridyl)ethylene (166). We felt that such compounds could be reduced to the desired indolylpyridylethanes by means of catalytic methods. This reaction sequence is outlined in (Scheme 51).

Scheme 51



1,1-Bis(3-indolyl)-1-(3-pyridyl)ethane (2) arises as a natural consequence of the mechanism for the condensation of indoles with carbonyl compounds.[119] Applied to this reaction it is as shown in (Scheme 52).

Scheme 52



(167)

The intermediate (167) formed in protic solvents is still subject to nucleophilic attack on the bridge head double bond, and in the presence of excess indole accepts another molecule to give the observed product (2).

1,1-Bis(3-indolyl)-1-(3-pyridyl)ethane (2) was prepared by a modification of the method employed by Bergman. This involves heating two molar equivalents of indole with one of 3-acetylpyridine in glacial acetic acid at reflux temperature for two hours. We then optimised the work up procedure using ice-cold aqueous ammonia (see experimental section p. 283), which reduced the severity of the conditions and improved the yield to 92%, which represents a 24% increase with respect to the original procedure.

1,1-Bis(3-indolyl)-1-(3-pyridyl)ethane (2) was thoroughly dried under reduced pressure, finely powdered and then vacuum pyrolysed at a temperature of 280-285^{o} for twenty minutes, in a system fitted with a nitrogen bleed. The pyrolysis was assumed to be complete when indole ceased to distill. The dark reaction mixture was cooled to room temperature and made acidic with hydrochloric acid. Extraction of residual indole with ether was followed by a base work up procedure, once again employing ice cold aqueous ammonia (p. 284), to give a sticky solid. Direct crystallization of this solid failed, from a variety of solvents, but T.L.C. showed it could be purified by chromatography on basic alumina, eluting with a mixture of ethyl acetate and light petroleum ether. The product from the column was then crystallized to give a white solid.

As this compound was not documented in the literature we carried out a full spectral characterisation. The mass spectrum exhibited a molecular ion of the desired mass to charge ratio $\underline{m/e}^{+\cdot}$ 220, and the ^{1}H n.m.r. spectrum displayed a pair of doublets in the region (5-6) p.p.m. corresponding to the protons of the bridge head double bond.

$\delta\left[(CD_3)_2SO\right]$ 5.50 and 5.81 (2 x 1H, 2 x d $\underline{J}$ 1.8Hz $\rangle C{=}{=}C\langle^{H}_{H}$)

The other physical data are entirely consistent with the structure (166), however, the yield of this pyrolysis step was a disappointing 10%.

Despite this difficulty, 1-(3-indolyl)-1-(3-pyridyl)ethylene (166) obtained in the previous experiment was dissolved in a mixture of ethanol and acetic acid, and the exocyclic double bond reduced by means of low pressure hydrogenation over a 10% palladium on carbon catalyst. After removal of the catalyst and extraction of the product with dichloromethane under basic conditions, this treatment gave an excellent yield (93%), of the desired 1,-(3-indolyl)-1-(3-pyridyl)ethane (113). Mass spectral analysis now showed, a molecular ion of mass to charge ratio $\underline{m/e}^{+\cdot}$ 222, and the ^{1}H n.m.r. spectrum displayed the expected doublet, quartet system corresponding to the bridge head CH_3CH group.

$\delta\left[(CD_3)_2SO\right]$ 1.60(3H, d, $C\underline{H}_3CH$), 4.30(1H, q, $CH_3C\underline{H}$)

The other physical data are entirely consistent with the structure (113) and with those obtained for a known sample of this compound prepared by the Grignard condensation of indolylmagnesium bromide and 3-(1-chloroethyl)pyridine[54].

We felt that this method offered some synthetic utility because steps A and C of (Scheme 51 , p, 120), are both highly productive, but the vacuum pyrolysis gave a poor yield. Accordingly we attempted to optimize the reaction conditions in the following way.

Firstly the temperature at which the pyrolysis is conducted was reduced to 250-260°, to minimise charring, but on working up this experiment in the original manner, approximately 20% by weight of starting material was obtained, and no significant increase in the yield of the desired product was achieved. The results of this experiment clearly show that relatively high temperatures are required to obtain a satisfactory yield of product.

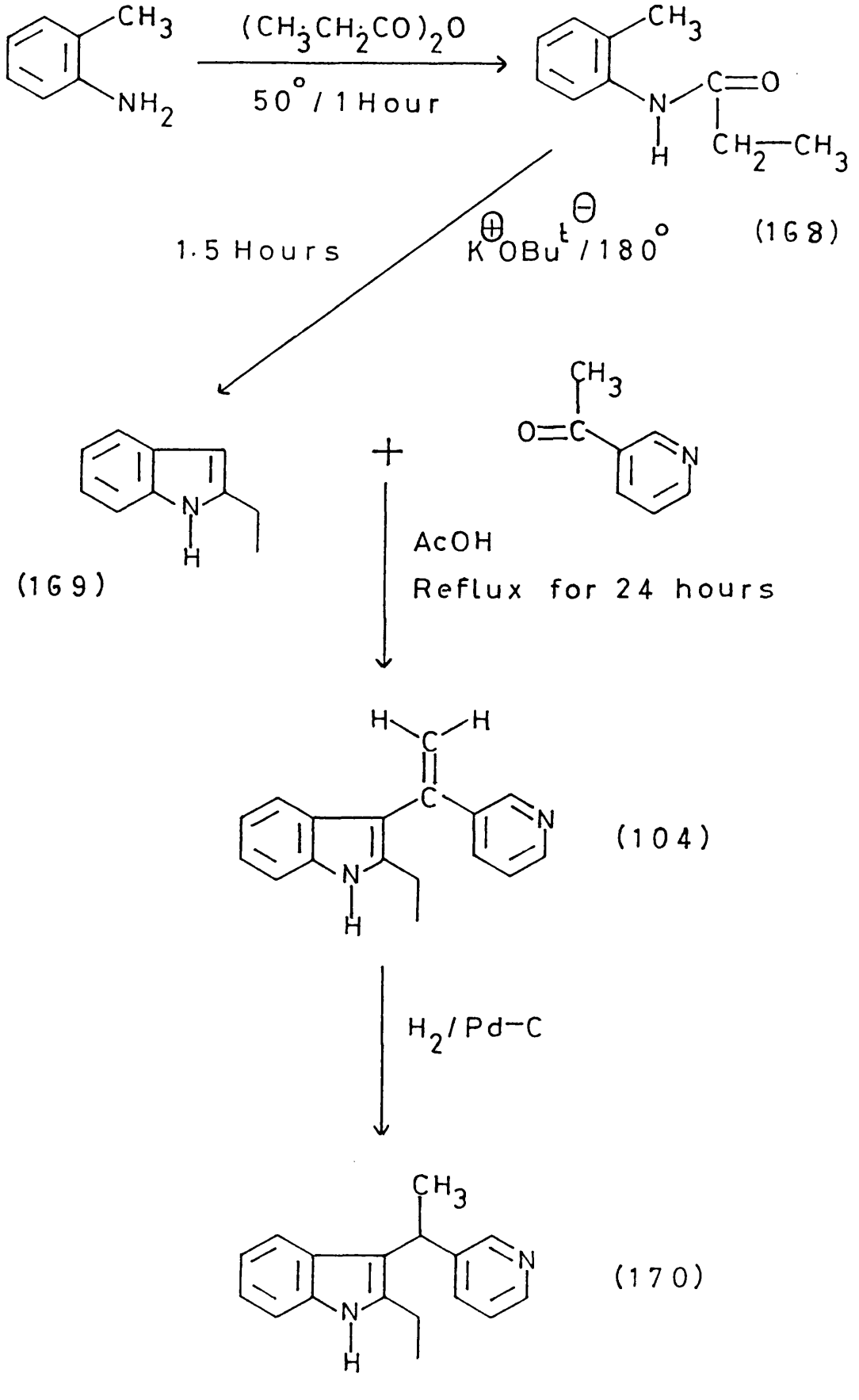
Next, the time taken for the pyrolysis was reduced to ten minutes at 350-360°, but the results of this experiment again indicated the reaction to be some 50% incomplete. No starting material had ever been isolated from the original reaction, where a pyrolysis time of twenty minutes was employed. It, therefore seemed unlikely that the poor yields obtained in this case, were due to an incomplete reaction, but in order to conclude this investigation, we carried out the following experiment.

The reaction was repeated using the original conditions, but this time extending the time taken for the pyrolysis to forty five minutes. However, on working up this experiment our expectations were confirmed, as the only extractable material proved to be a dark, resinous gum, which failed to yield to the chromatographic techniques applied successfully before.

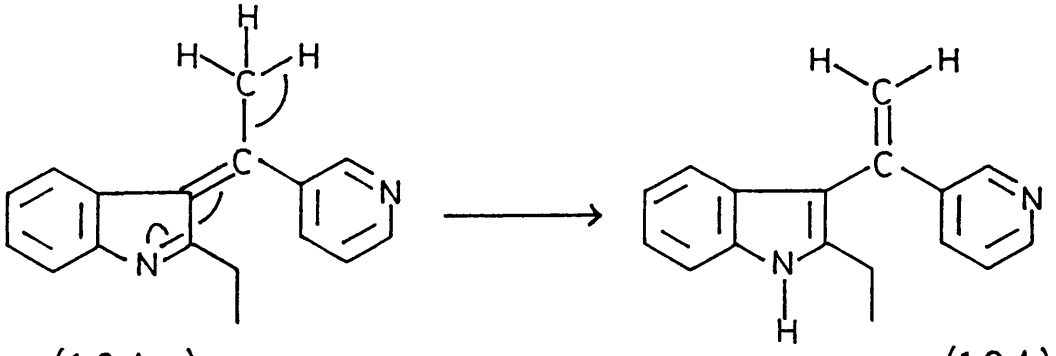
In view of these difficulties it was felt that this method could not be usefully applied in a case such as the one under consideration, where the indole to be used was a costly substituted compound obtained after many weeks of labour. It is apparent from this investigation that a great deal of experience is required in operating such vacuum pyrolysis experiments in order to obtain similar results to those found by Swedish Chemists[118]. Subsequent discussion with the original workers, also revealed that the heat source in their experiments was a costly, thermostatically controlled, fluidized bed, a point not clearly defined in their communication. This apparatus was not available to us, and since it does offer a more controlled and even method of heating than the more conventional methods available in this laboratory it is possible that this contributed to the low yields we obtained for the pyrolysis step, in this otherwise promising route.

A second, interesting synthesis[6] carried out in Sweden at this time was that outlined in (Scheme 53).

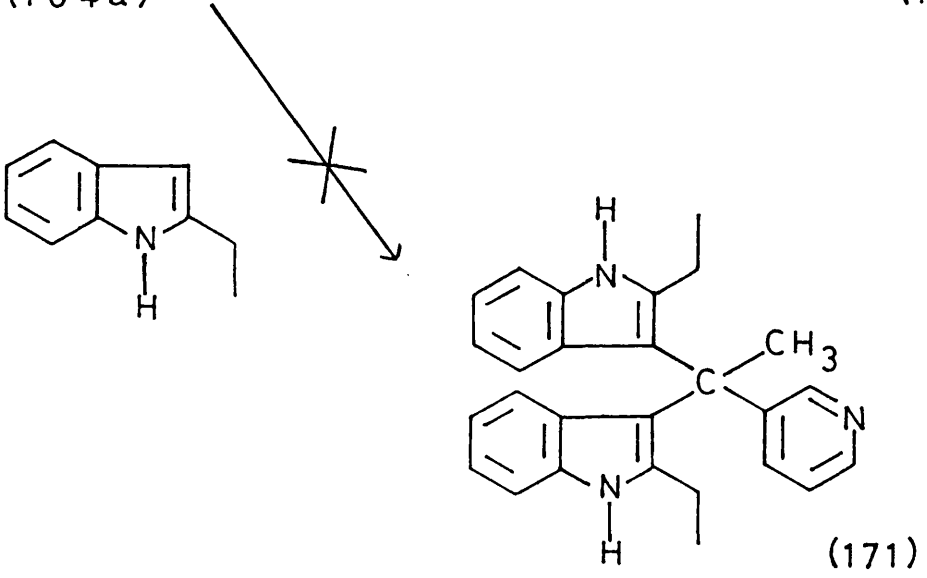
Scheme 53.


CH3
NH2
(CH3CH2CO)2O
50° / 1 Hour
CH3
N
H
C=O
CH2—CH3
(168)
1.5 Hours
K⊕ OBut ⊖ / 180°
(169)
+
CH3
O=C
N
AcOH
Reflux for 24 hours
H
H
C
C
N
N
H
(104)
H2 / Pd-C
CH3
N
N
H
(170)

This work involved the preparation of 2-ethylindole by condensation of o-toluidine and propionic annhydride to give 2-methylpropanilide (168), which was cyclized with potassium tertiary butoxide at high temperature in a Madelung procedure. Subsequent condensation of 2-ethylindole(169) with 3-acetylpyridine by heating at reflux temperature in glacial acetic acid afforded the 1:1 product (104). The explanation for this mono-condensation is considered[6] to be the steric hindrance due to the bulky ethyl group, which prevents further attack on the intermediate (104a) by a second molecule ot 2-ethylindole and hence formation of the Bis diethylindolyl compound (171). The resulting 1-(2-ethyl-3-indolyl)-1-(3-pyridyl)ethylene (104), was easily reduced to its saturated analogue (170), with hydrogen over palladium on carbon.

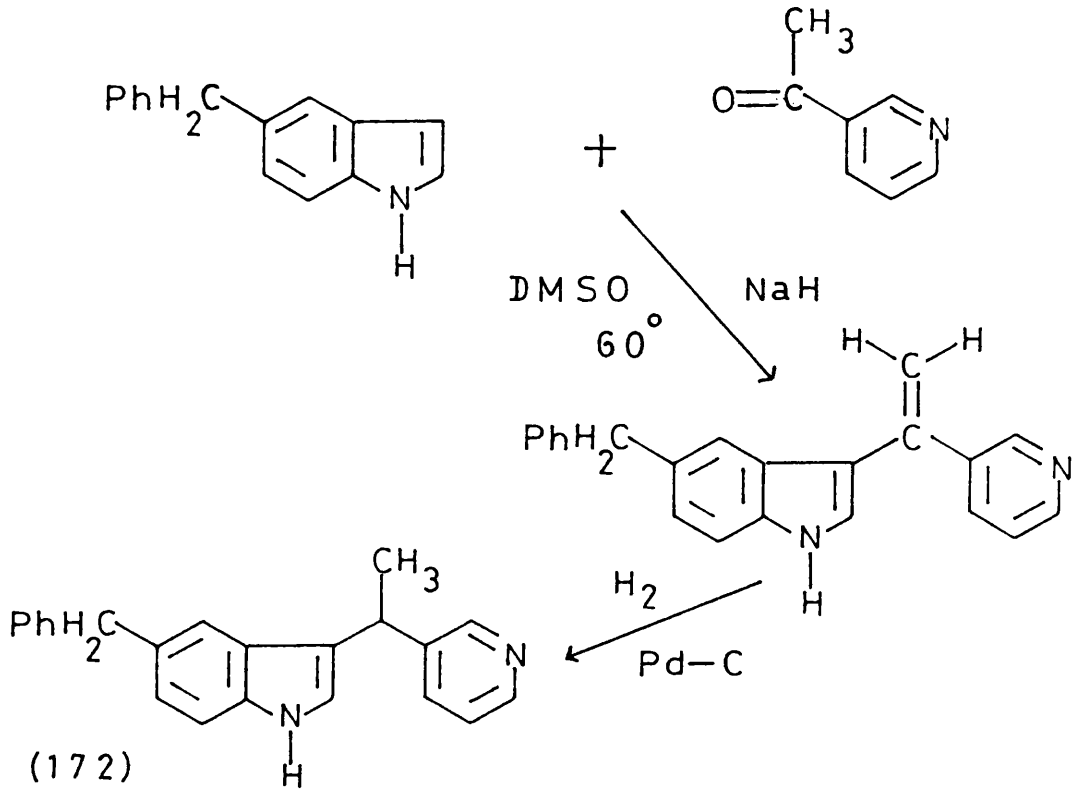


(104a) (104)



(171)

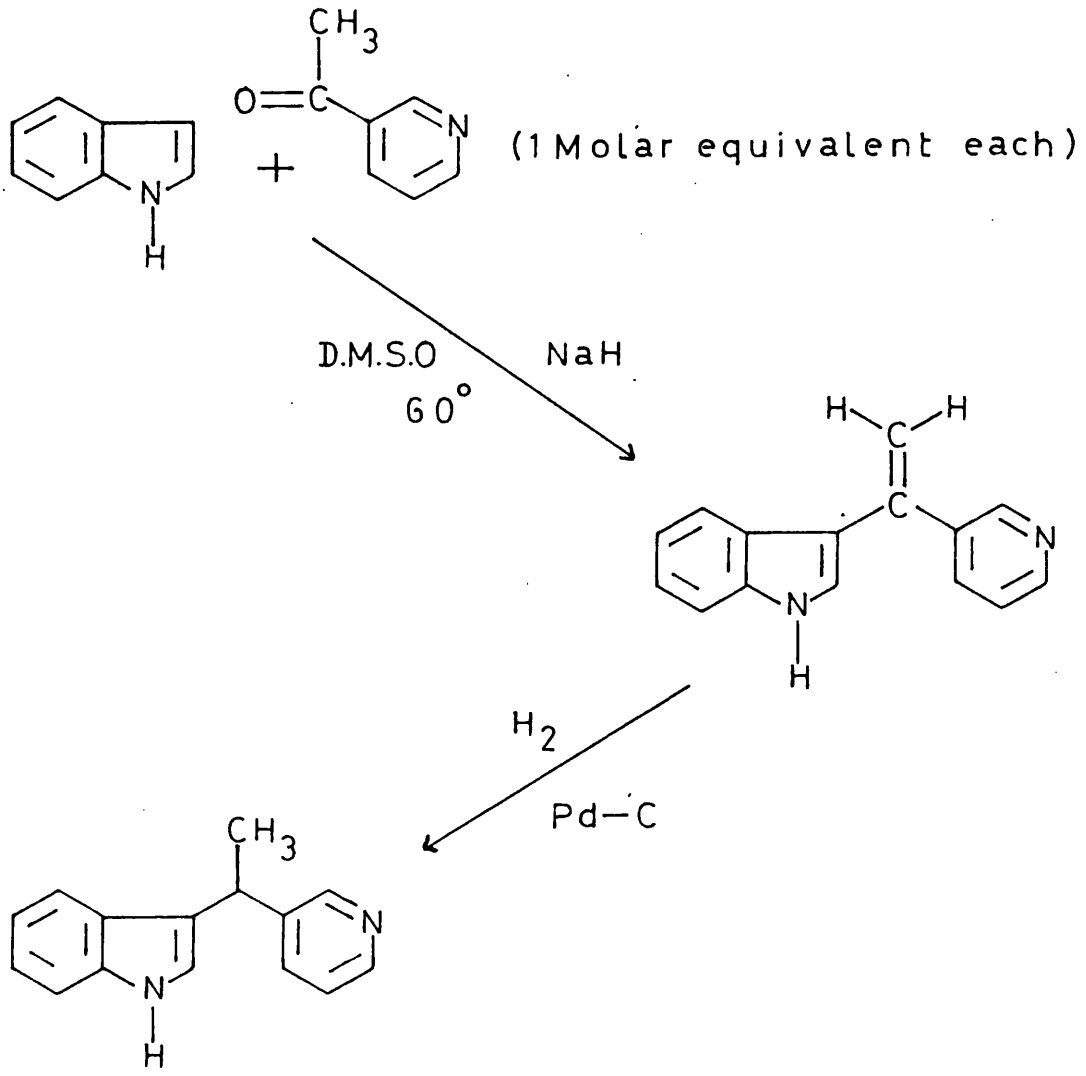
The work described above, although attractive in its simplicity was of no direct use to us because our method of choice for the synthesis of 8-hydroxyellipticine (see, p. 62) precludes the use of any indole 2-substituent that cannot be easily removed at a later stage. A literature search for such an easily removable, bulky blocking group, that could be introduced or built into the 2-position of 6-methoxyindole, failed to reveal any useful procedures. However, at about this time we received a second unpublished communication from Sweden[118], indicating that the synthetic utility of the mono-condensation might not be restricted to indoles bearing a large substituent in the 2-position. This work involved the 1:1 condensation of 5-benzyl-indole with 3-acetylpyridine using sodium hydride in dimethyl-sulphoxide at 60°, to give the appropriate indolylpyridylethylene, which can be catalytically reduced to the corresponding indolyl-pyridylethane (172) as shown.



(172)

As this indole also contains a bulky group, we considered whether this is of significance, but it does not appear to be so orientated that it should block formation of the appropriate bis-diindolyl compound. This lead us to speculate that the success of this condensation might be attributed to the reaction conditions, rather than the steric constraints in the starting materials. Accordingly we decided to carry out a similar experiment using indole itself and 3-acetylpyridine. This projected route to the desired indolylpyridylethanes is outlined in (Scheme 54).

Scheme 54



Due to the fact that this work was unpublished we lacked precise experimental details. Therefore, we set out to establish the best method of performing this reaction, and carried out several experiments under a variety of conditions as follows:

Firstly, equimolecular quantities of the two reactants in dry dimethylsulphoxide were treated with two molar equivalents of sodium hydride at 60^{o}. After three hours the excess sodium hydride was destroyed with ethanol, the ethanol evaporated, and the residue poured into an excess of water. This was extracted, first with ether to remove starting materials and then with dichloromethane to obtain the product. However, the dichloromethane extract gave a dark and sticky gum which would not yield to the usual chromatographic procedures.

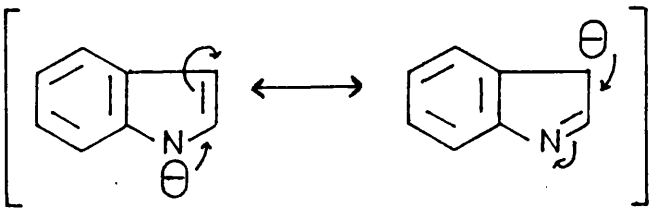
We felt that the failure to isolate the product of this reaction might be due to its solvation by dimethylsulphoxide in the aqueous phase. Therefore, we modified the work-up procedure as follows.

The experiment was carried out as before except that all the solvents were evaporated under vacuum, prior to extraction. T.L.C. analysis of the dichloromethane extract on basic alumina plates eluting with a mixture of ethyl acetate and light petroleum ether showed it to consist of a multicomponent mixture of high polarity compounds. None of the desired product could be detected when compared with an authentic sample of 1-(3-indolyl)-1-(3-pyridyl) ethane, but a high proportion of unreacted starting materials were isolated from the ether extracts.

These results caused us to consider the possibility that the reaction conditions were not sufficiently energetic, thus, we carried out two further experiments.

Firstly, the reaction was repeated as before, except that the temperature was held at 60° for sixteen hours. This treatment gave little starting material, but much polymeric gum. Once again no product could be detected by T.L.C. Finally, the experiment was repeated at a temperature of 120°, for only two hours, but as before a polymeric gum was obtained that would not yield to chromatographic techniques.

The failure of this reaction lead us to speculate on its possible mechanism. Presumably the strong base N-deprotonates indole to give the resonance stabilized indolyl anion (173).



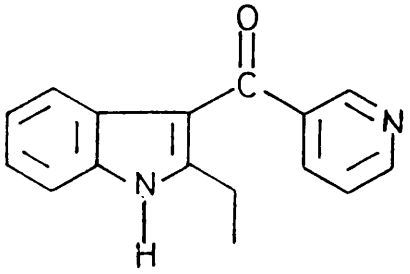
(173)

This anion can then react with electrophiles either on the nitrogen or at the C-3 position. Joule and Smith[120] comment that the ratio of N- to C-3 substitution by an electrophile depends upon a number of factors. These include the nature of the metal cation, the type of indole substituents already present, if any, and their positions. The type of solvent used, the reaction temperature and the nature of the electrophile. If the cation is a

sodium ion, the reaction is thought to occur predominately at the nitrogen atom.

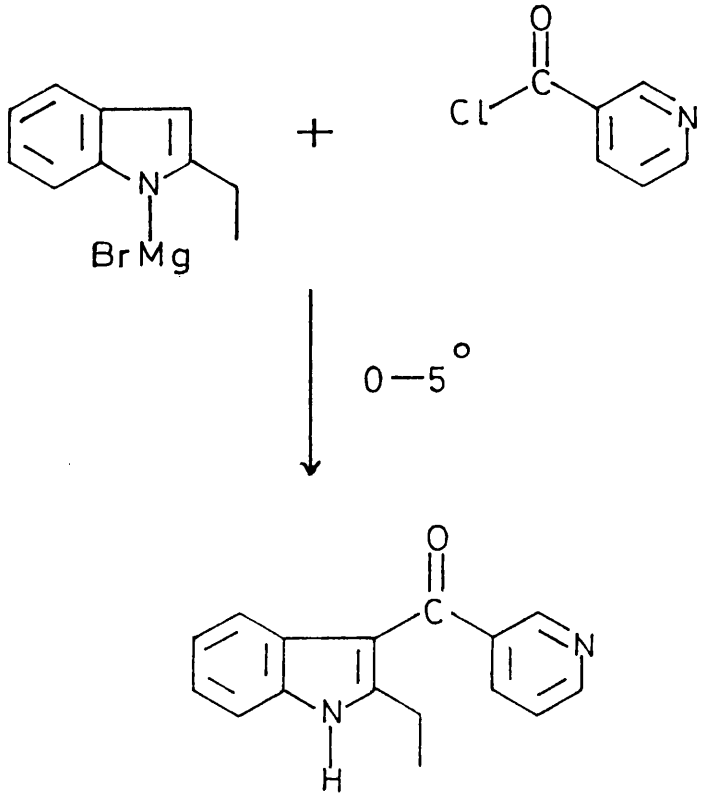
As neither product was isolated from any of the reactions between indole and 3-acetylpyridine we assume that the reaction involving 3-benzylindole is, in some way favoured with respect to indole itself. However, this is difficult to justify on electronic or steric grounds and we cannot offer a precise rationalization of the failure of this reaction at the present time.

At this point we felt that it would be unproductive to continue with further attempts to achieve this reaction and we turned our attention to a photochemical study of the compound (174).



(174)

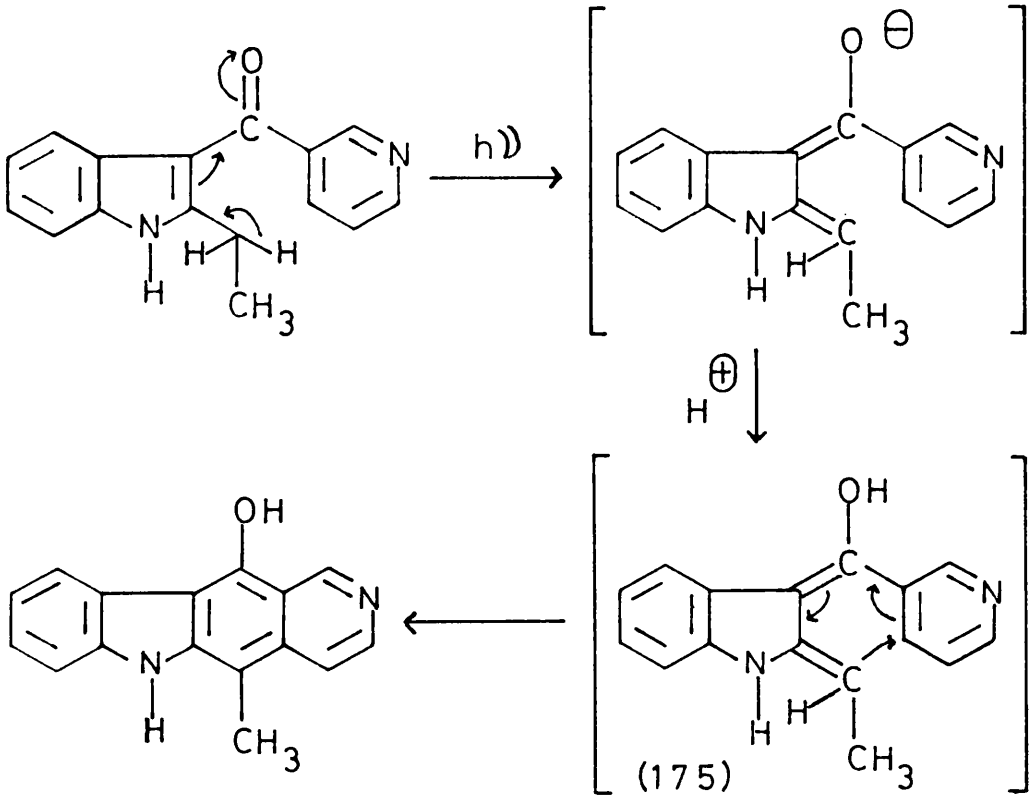
This compound was prepared from 2-ethylindolylmagnesium bromide and nicotinyl chloride as shown.[121]



It is interesting to note in passing that the yield of (174) was 72%, which further confirmed our suspicions that the low yields obtained with the Grignard reaction discussed earlier on (p. 117) are due to the instability of the haloethylpyridines employed, rather than problems associated with the formation of the indolylmagnesium bromides.

Having the compound (174) in hand we envisaged the possibility of a direct photochemical ring closure, to give the pyrido[4,3-b]carbazole system via the trienol intermediate (175), as shown in (Scheme 55).

Scheme 55.

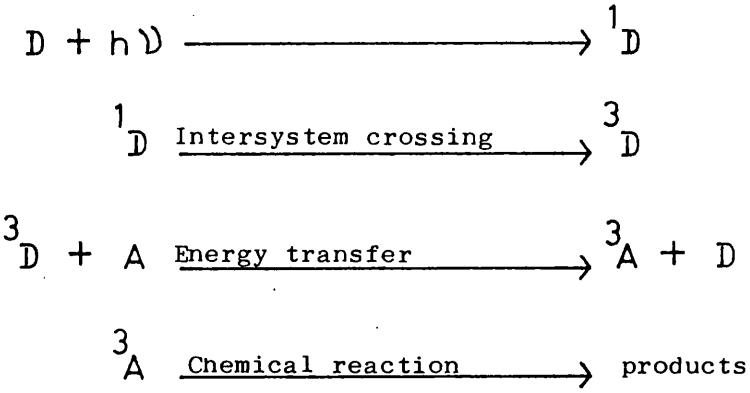


The photochemical experiments involving (174) were carried out in methanol using ultraviolet light generated by a high pressure source, (see experimental section, p. 295). The progress of the reaction was monitored by ultraviolet spectroscopy. The pyrido[4,3-b]carbazole system has a characteristic electronic absorption spectrum displaying a complex of strong bands close to 300 nm. The starting material (174) has absorption maxima at 238 and 328 nm, thus the change on reaction would be obvious. The reaction was examined every two hours for sixteen hours, but no change was observed.

Further consideration of this reaction caused us to speculate that it might proceed through a low energy triplet state. An

attempt was made to increase the concentration of such a triplet state by including a small quantity of benzophenone in the reaction mixture. Benzophenone is a highly efficient triplet sensitizer[122,123], forming a high concentration of its triplet state by intersystem crossing and then transferring this energy to the substrate molecule as outlined in Fig. 4.

Fig. 4.



Where D = Donor ≡ benzophenone, A = Acceptor ground state ≡ in this case to (174) and the superscripts 1 and 3 refer to singlet and triplet states respectively.

When this reaction was carried out, no change occurred for eighteen hours and then a shift to shorter wavelength was observed. After twenty hours the change was complete and showed one band with an absorption maximum of 256 nm. T.L.C. analysis confirmed that no pyrido[4,3-b]carbazoles were present and no fluorescence under ultraviolet light could be detected. The mixture consisted of seven components, four major and three minor. Removal of the solvent and mass spectral examination of the residue confirmed this, showing four major

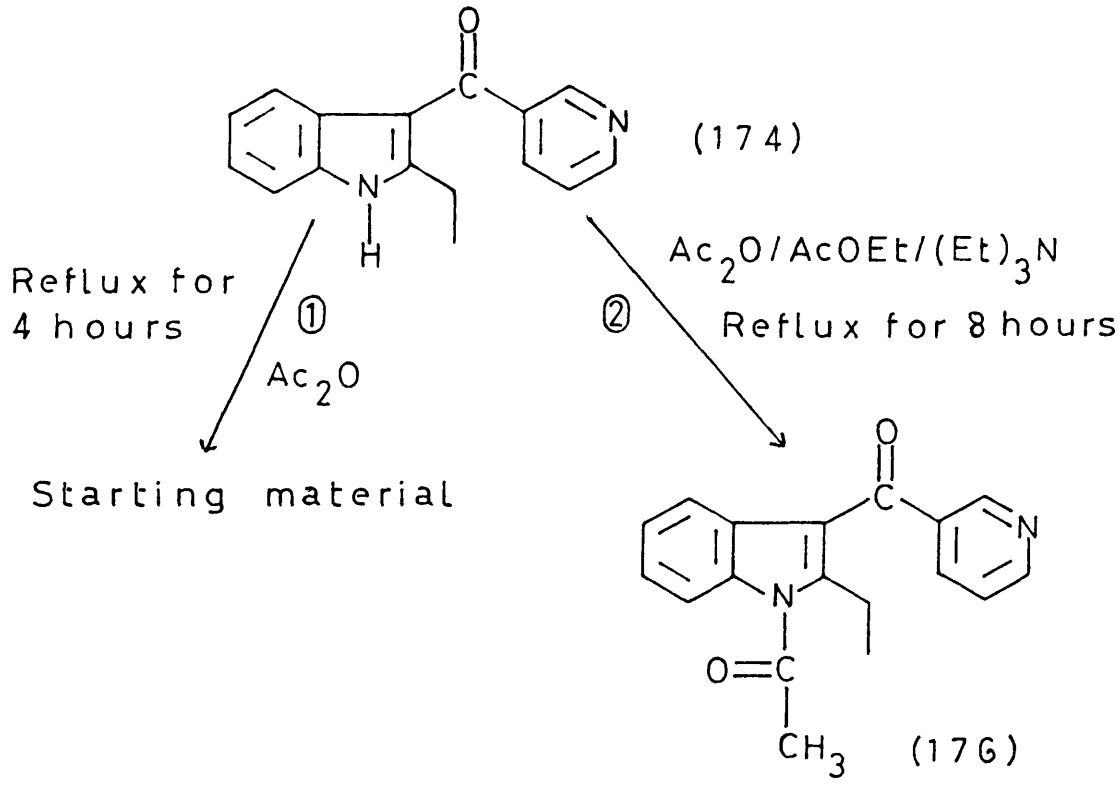
molecular ions at mass to charge ratios of $\underline{m/e}^{+\cdot}$ 223, 193, 137 and 123.

It was clear from these investigations that the molecule (174) had simply fragmented due to prolonged exposure to ultraviolet light. Further examination of this mixture was not undertaken because our main objective was to obtain an efficient route to ellipticines.

We now turned our attention to modifying the compound (174) in such a way, that it would become a better substrate for photochemical cyclization. Experience with the chemistry of these compounds has led us to regard them as vinylogous amides rather than ketones, thus the carbonyl stretching frequency is at 1595 cm^{-1} in the infrared spectrum. In view of the fact that many ketones readily undergo photochemical reactions it was felt that if the carbonyl group of (174) could be made more ketonic in nature it would be more likely to undergo the desired reaction.

To this end, we attempted to $N_{(a)}$-acetylate (174) prior to carrying out the photochemical experiment. However, this proved surprisingly difficult, and the reactions outlined in (Scheme 56), were carried out.

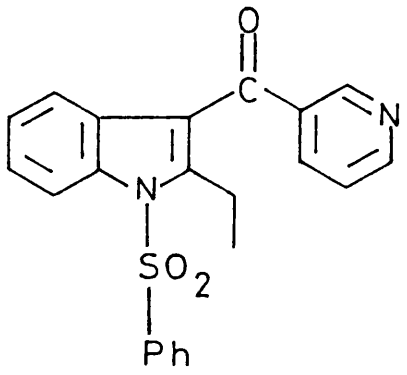
Scheme 56



Firstly the acetylation was carried out in excess acetic anhydride at reflux temperature, but this returned only starting material. The reaction was then carried out under more vigorous conditions, using a mixture of acetic anhydride, ethyl acetate and triethylamine at reflux temperature for eight hours. When this reaction was worked up the desired product (176) was obtained as colourless plates in 87% yield. T L C of the reaction mixture at periods of less than eight hours, showed it to consist of a mixture of starting materials and product, indicating that the compound (174) is much more difficult to acetylate than might be expected.

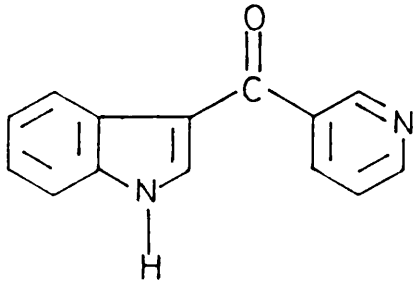
We noted that the carbonyl frequency of (176) was at ν_{max} 1640 cm^{-1}. Thus, the photochemical experiment was again carried out using benzophenone as the sensitizer, but unfortunately this gave similar negative results.

Finally in a bid to reduce still further the amidic character of the substrate (174), we attempted to form the N-benzenesulphonyl derivative (177).



(177)

Firstly the compound (174) was dissolved in ethyl acetate and treated with benzenesulphonyl chloride in the presence of triethylamine at the reflux temperature, but T.L.C. analysis showed only starting materials to be present. Secondly, (174) in dry dimethylsulphoxide was treated with sodium hydride in the presence of an equimolar quantity of benzenesulphonyl chloride at 40^{o}.[124] However, T.L.C. and mass spectral analysis, once again, indicated that only starting materials were present. These frustrating results were rationalized after further work in this laboratory with the compound (178).[125]

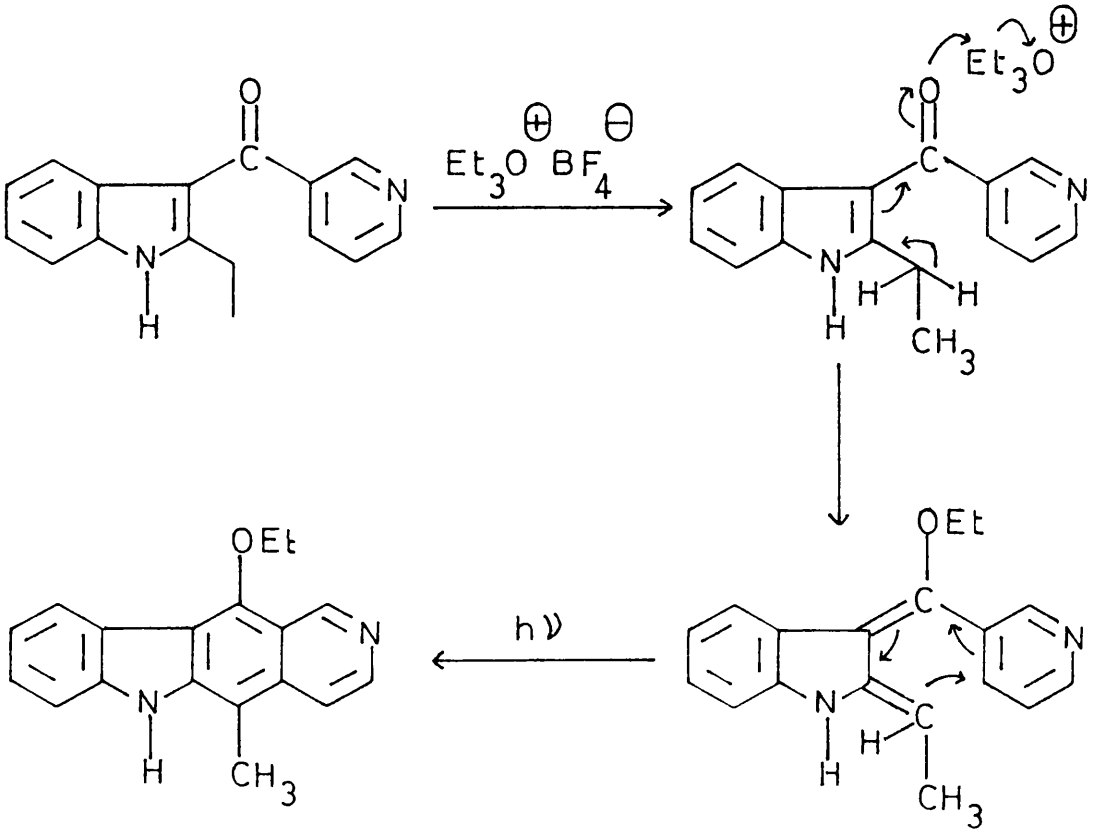


(178)

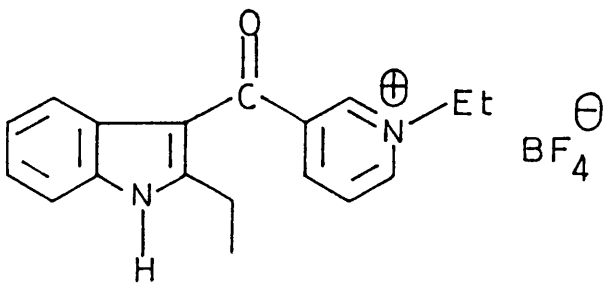
This compound, lacking the 2-ethyl group was found to undergo smooth acetylation by the method unsuccessfully applied to the compound (174), and to form the N-benzenesulphonyl analogue. It is apparent from these results that steric hindrance of the N–H group by the adjacent bulky ethyl function is the main cause of these problems. Hence the small acetylium ion $CH_3C{\equiv}O^+$ derived from acetic annhydride[126] does attack with difficulty, but the larger benzenesulphonyl function is inhibited.

In a final attempt to achieve the desired photochemical cyclization of compound (174), and in view of the previous difficulties we decided to employ a different mode of attack using triethyloxonium tetrafluoroborate. This substance will ethylate amidic carbonyl groups[127] to give the appropriate vinylogous ethers. We then envisaged the reaction sequence shown in (Scheme 57).

Scheme 57.



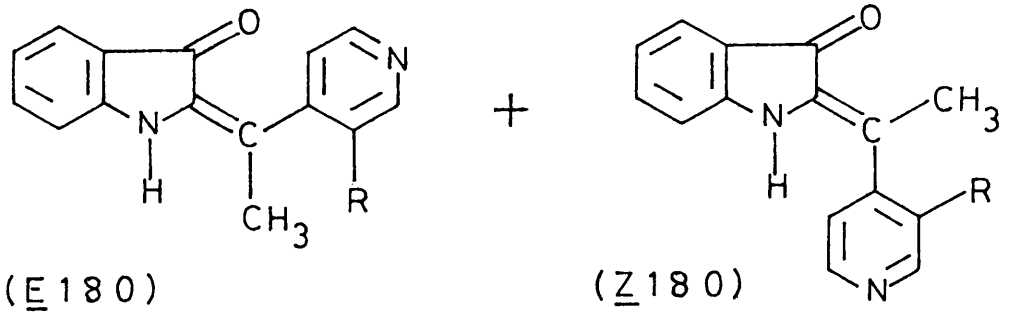
We treated the compound (174) with <u>two</u> molar equivalents of Meerwein's reagent because of the possibility of a competing reaction with the pyridine nitrogen. The reaction was carried out under anhydrous conditions in dichloromethane at room temperature. However, the sole product of this reaction was the ethyl tetrafluoroborate salt (179).



(179)

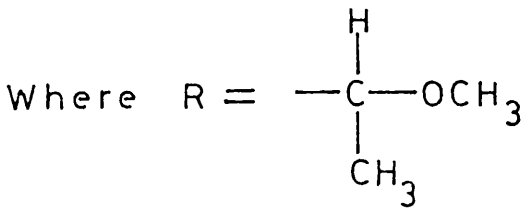
This result clearly shows that the pyridine nitrogen is indeed more reactive to the triethyloxonium tetrafluoroborate than the carbonyl function. Further reaction of the carbonyl group in the salt with excess Meerwein's reagent is presumably prevented by the insolubility of this salt in dichloromethane.

Before discontinuing the photochemical approach to the tetracyclic skeleton we considered other ways of generating the desired triene system. Some years ago in this laboratory[18], it became necessary to synthesize the E- and Z-forms of 2-[1-(4-pyridyl)ethylidene]indolin-3-ones of the type (180), see (p. 18).

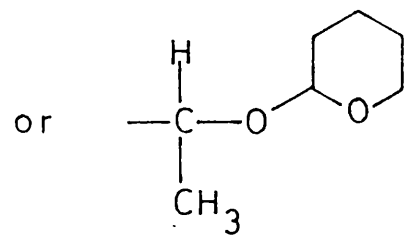


(E 180) + (Z 180)

Where R =

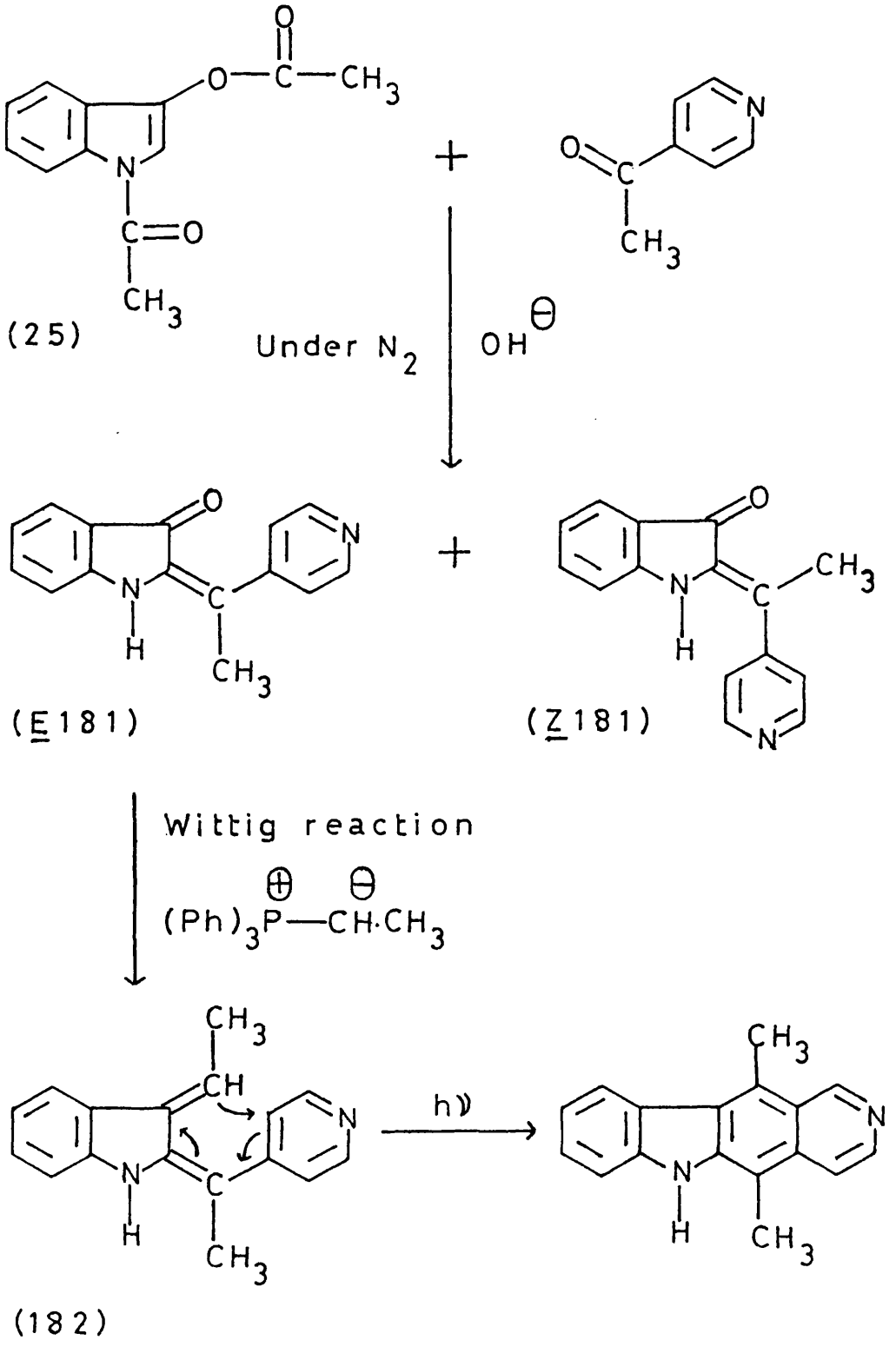


or



We now considered the possibility of synthesizing compounds of this type lacking the R group, which might then be employed in the reaction sequence outlined in (Scheme 58).

Scheme 58.



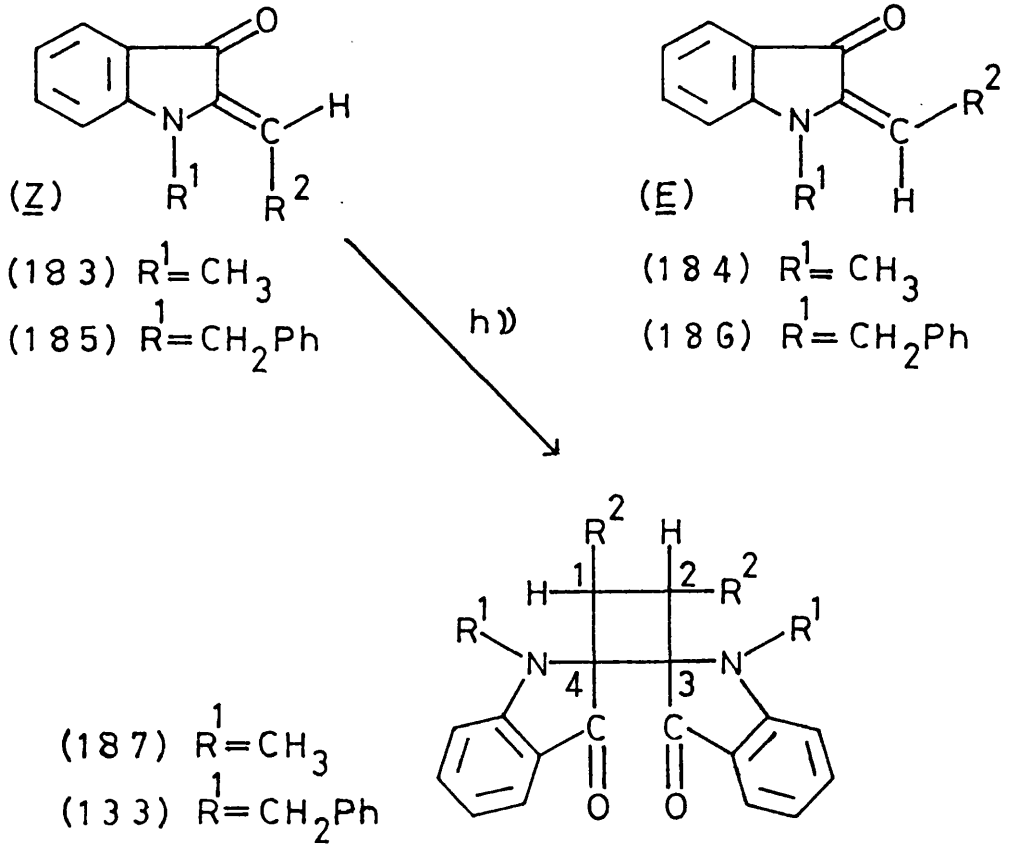
Our initial aim was to separate the E- and Z-forms of (181) and convert the carbonyl group of the E-form to the ethylidine function by means of a Wittig reaction using the ylide generated from triphenylethylphosphonium bromide and butyl lithium. The resulting masked triene system (182), might, then photolyse to give ellipticine. However, this procedure presented a number of most frustrating difficulties.

1-Acetyl-3-acetoxyindole (25) was condensed with 4-acetylpyridine in the presence of aqueous alcoholic sodium hydroxide under a protective nitrogen atmosphere to give the mixed 2-pyridylmethyleneindolin-3-ones (181 E- and Z), in 88% yield. It was then necessary to separate these isomers, but this proved much more difficult than expected. T.L.C. analysis of the mixture on basic alumina, neutral alumina and silica, using a variety of solvent systems all showed six components, two red and the remainder yellow in colour. However, the mass spectrum of the mixed isomers (181 E- and Z-) indicated that only one molecular ion was present, at the expected mass to charge ratio of $m/e^{+\cdot}$ 236.

The question arose, were we dealing with high molecular weight, relatively involatile impurities that did not show up in the mass spectrum, or alternatively, were further reactions occurring on the chromatographic supports. Mass spectrometry at elevated temperatures failed to show any other molecular ions, which tended to indicate that the second possibility was the correct one. Analysis and separation by gas chromatography was rejected because the 2-pyridylmethyleneindolin-3-ones are very polar compounds with

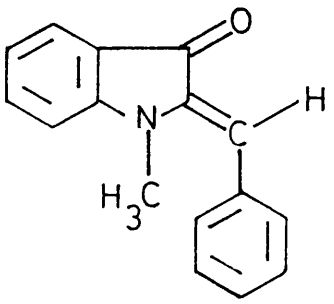
long retention times and doubtful thermal stabilities. We did not have an efficient high pressure liquid chromatograph available to us at this time, so we resorted to a literature search for compounds of the type (181). This eventually revealed a paper by Hooper and Pitkethly[128] in which they reported the isolation of cyclobutane derivatives as impurities in the preparation of E- and Z-forms of N-alkyl-2-arylmethyleneindolin-3-ones[129] as outlined in (Scheme 59).

Scheme 59.



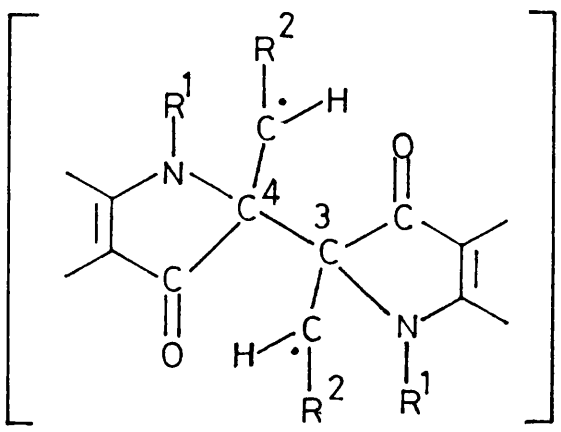
a; R^2 = Ph
b; R^2 = 2-ClC_6H_4
c; R^2 = 4-ClC_6H_4
d; R^2 = 2-$CH_3OC_6H_4$
e; R^2 = 4-$CH_3OC_6H_4$
f; R^2 = 4-$(CH_3)_2NC_6H_4$
g; R^2 = 2-$O_2NC_6H_4$
h; R^2 = 4-$O_2NC_6H_4$
i; R^2 = 4-$HO_2CC_6H_4$
j; R^2 = 2,6-$Cl_2C_6H_3$

It was noted that when N-methyl-2-phenylmethyleneindolin-3-one was prepared in the absence of light, at 0^{o} or at reflux temperature only the orange Z-isomer (183a) was obtained.



(183a)

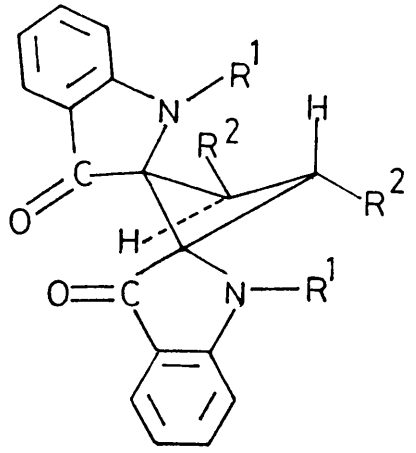
However, irradiation of solutions containing (183a) gave the same yellow cyclobutane derivatives as those occurring in the synthesis of the N-alkyl-2-arylmethyleneindolin-3-ones that were not protected from light. The yellow cyclobutane derivatives arise through a photodimerisation reaction, and the authors proved the structures of compounds (183-188), by means of a detailed proton n.m.r. examination combined with mass spectral and analytical data. Photodimerisations of this sort have been reported before[130,131], and are characteristic of $\alpha\beta$-unsaturated carbonyl compounds. The authors indicate that the reaction probably proceeds through an intermediate biradical triplet state such as that shown.[132]



The influence of substituents in the aryl ring of the indolinone on the ease and extent of cyclobutane formation is known[133] to be in accordance with the formation of such an intermediate. Hooper and Pitkethly also comment that the nature of the aryl substituent (R^2) of the indolinone influences the formation of the cyclobutanes. When the substituent (R^2) was strongly electron withdrawing (183 g-j) and (185 h), no cyclobutanes were obtained. As (R^2) becomes more electron releasing the yields of cyclobutanes increase. Indeed, in one-pot reactions carried out to prepare (187 e and f) dimerisation was facilitated to such an extent that indolinones could not be isolated.

When the cyclobutane (187; R^2 = Ph), derived from (183a), was heated in the absence of light at a temperature in excess of 130°, equal quantities of Z- and E-N-methylindolinones (183a and 184a) were isolated in very high yield. This thermal cycloreversion reaction may proceed by either a radical or a concerted process. The concerted mechanism would proceed by the disfavoured and rare suprafacial-antarafacial $\left[{}_2{}_s + {}_2{}_a \right]$ mechanism. If the concerted mechanism did occur it would be expected to be stereospecific and give either an all E- or an all Z- product. The observed 50% yield of each isomer would tend to indicate a 1,4-diradical intermediate in which bond rotation has reached equilibrium before cleavage. A number of papers agree that cycloreversions of cyclobutanes involve radical intermediates,[134,135] and satisfactory experimental proof for $\left[{}_2{}_s + {}_2{}_a \right]$ cycloreversions is lacking[136,137].

However, Hooper and Pitkethly, speculate that the rigid cyclobutane structure (189), in which there is considerable twisting of the C_2 - C_3 bond would facilitate the concerted process which requires a 180^{o} rotation of one of the terminal atoms.



(189)

It is not fully established which of these theories is the correct one, and as thermal cycloreversions of cyclobutanes may proceed by either a radical or a concerted mechanism[138], it could well be that these are competing pathways.

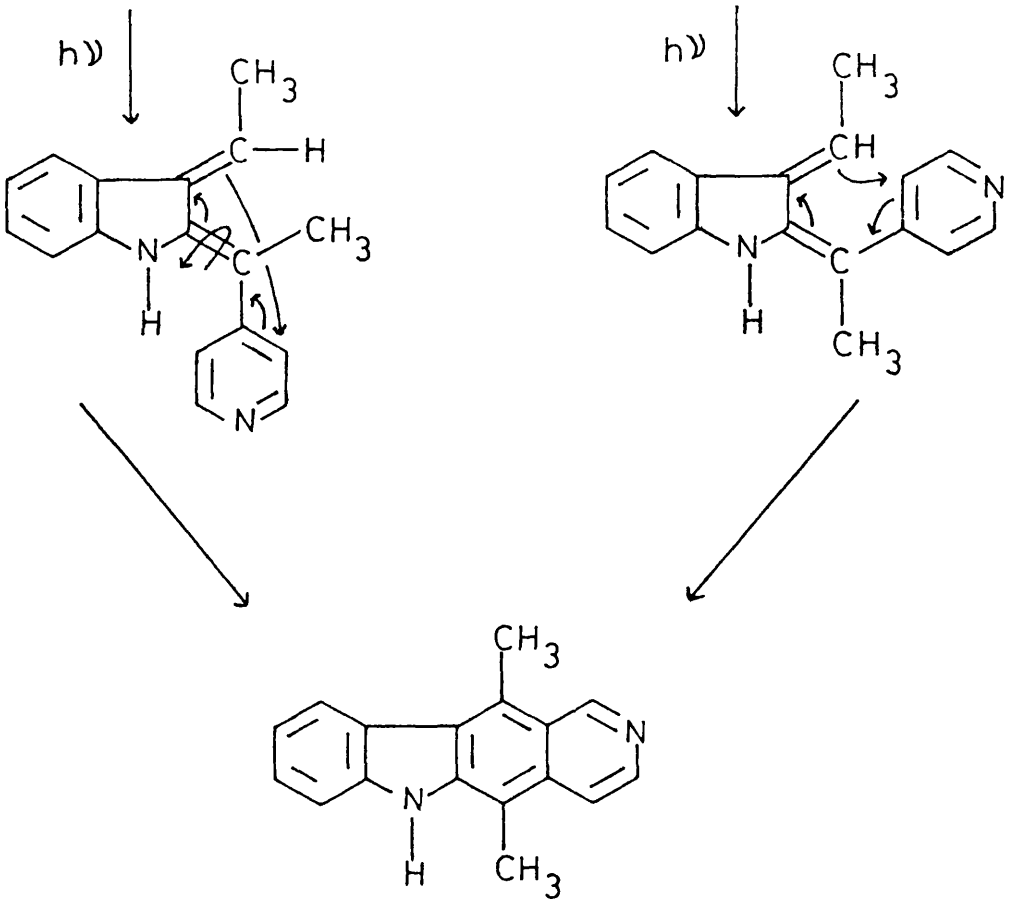
Interestingly, Hooper and Pitkethly comment that N-unsubstituted arylmethyleneindolin-3-ones did not photodimerise. At first sight our results would appear to be in conflict with this, but the indolinones (181; E- and Z-), only gave further products when exposed to chromatographic support materials. T.L.C. analysis was again carried out but this time under conditions of reduced illumination, this showed that the reaction still occurred on alumina but to a much decreased extent on silica. It would appear from these results that chromatographic supports act as a

catalyst for this reaction. The indolinones (181; E and Z) have the vinyl hydrogen atom of (183a), replaced by a bulky methyl group which would tend to inhibit photodimerisation via a 1,4-diradical intermediate, as Pitkethly[139] has shown that such intermediates are sensitive to steric factors. Thus, when indolinones in which the vinyl hydrogen atom has been replaced by a phenyl group or a bromine atom are exposed to light, no dimerisation can be observed. In view of these considerations we concluded that the indolinones (181; E- and Z), may be constrained on the alumina surface in such a way that steric crowding is relieved to some extent and the reaction can proceed more readily. Furthermore, similar indolinones (26, 27, 44 and 45, see; pp. 18 and 26), have been prepared in this laboratory at earlier times[16,18], by methods not involving chromatography, and in these cases no photoinduced further products could be detected. These reactions would, we felt, form an interesting study in their own right, but as our primary concern was to develop an efficient route to the ellipticine skeleton we lacked the necessary time to pursue these enquiries.

Despite these difficulties we decided to attempt the separation of the E- and Z-forms of (181). We required as short a column contact time as possible, so it was decided to use the technique of "flash" chromatography. This was carried out on silica gel in the absence of light, using a positive nitrogen pressure, and eluting with a mixture of dichloromethane and methanol. In this way a small amount of the desired E-isomer of (181) was isolated from the solvent front, but the difference in R_f values between the two forms was insufficient to allow a separation in quantity.

In view of these separation problems, and hence, the small amount of the E-form of (181) obtained, we decided to continue to the next stage of the synthesis with the mixed E- and Z-forms of (181). This we realized would in turn give rise to both the E- and Z-forms of the masked triene system (182, Scheme 58; p. 142), as shown in (Scheme 60). However, in the photochemical step, free rotation of the carbon-carbon single bond, formed at the C-2 position of the indoline moiety in the Z-isomer, may occur and for this reason it was felt that separation of the isomers was not obligatory.

Scheme 60.



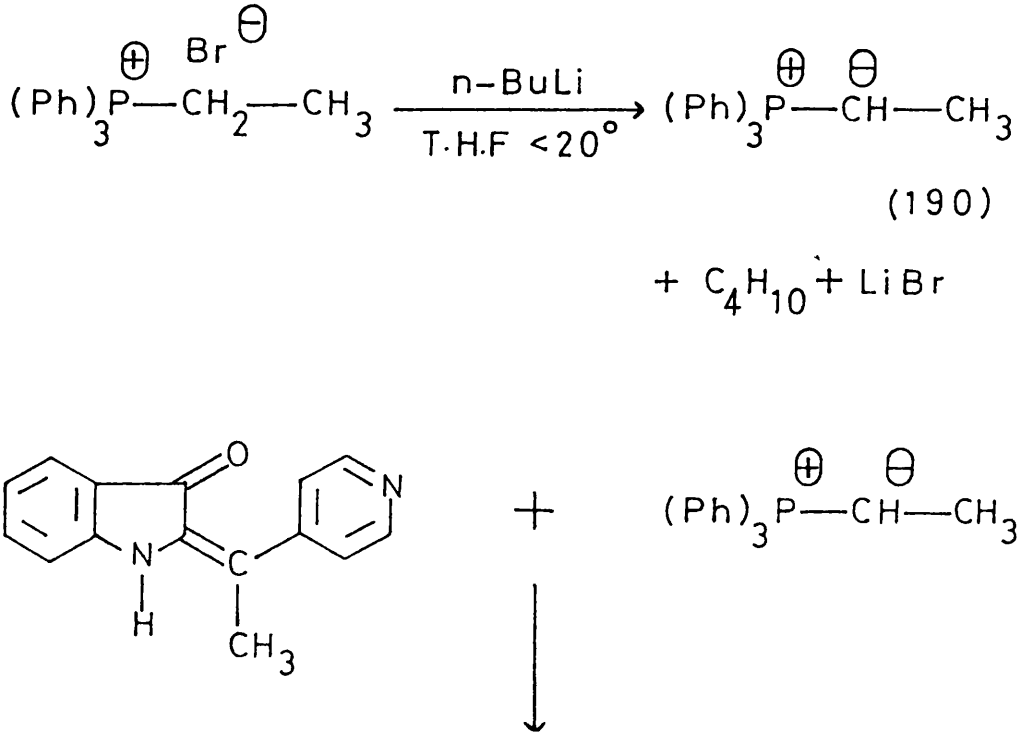
Where electron pairs move before bond rotation.

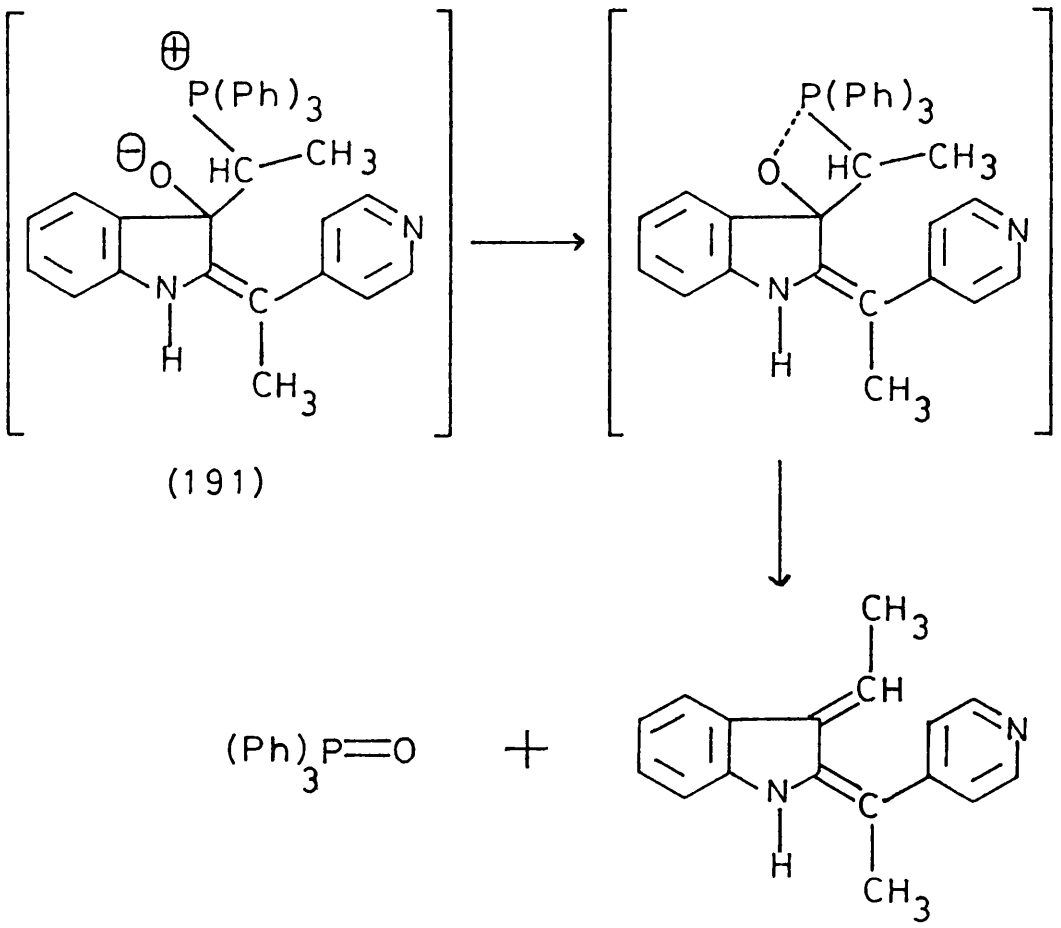
Firstly we prepared triphenylethylphosphonium bromide[140] by the reaction of triphenylphosphine and ethyl bromide in dry benzene at 0°. Stirring overnight gave a 92% yield of this compound.

$$(Ph)_3P + CH_3CH_2Br \longrightarrow (Ph)_3P^{+}-CH_2-CH_3\ \ Br^{-}$$

In order to carry out the next stage of the synthesis it was necessary to find a suitable solvent. The extensive review by Wadsworth,[141] indicates that the majority of Wittig reactions are carried out in diethyl ether or 1,2-dimethoxyethane, but solubility tests showed that the indolinones (181, E- and Z) are almost insoluble in these solvents. We were, therefore obliged to resort to tetrahydrofuran T.H.F. as the next most suitable solvent. The formation of the ylide (190), and its reaction with the indolinones (181; E- and Z, only one is shown), was performed as a one-pot reaction, using butyl lithium as the base. This sequence is outlined in (Scheme 61).

Scheme 61





(191)

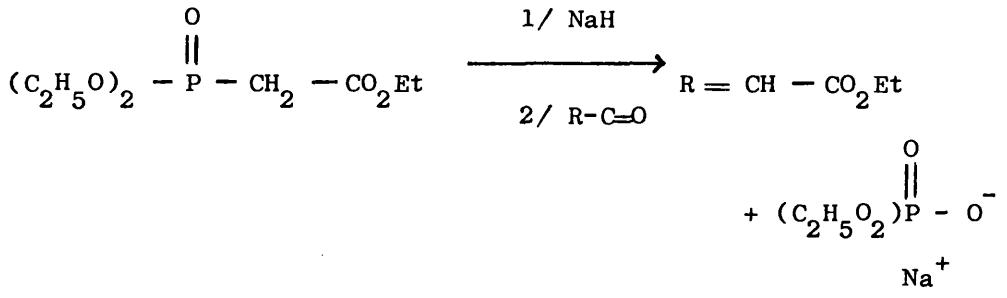
Equimolar quantities of triphenylethylphosphonium bromide and butyl lithium were reacted under anhydrous conditions in T.H.F. One molar equivalent of the indolinones (181, E- and Z), were then added and the reaction stirred at room temperature for a period of four hours.

Examination of the chloroform extract from this reaction showed that the carbonyl band in the infrared spectrum was now absent, but the ^{1}H n.m.r. spectrum displayed too large a number of aromatic protons, indicating that the triphenyl group was still present. We felt that this could be rationalized in one of two possible ways. Firstly, the product might be contaminated by

triphenylphosphine oxide due to the use of T.H.F. as the solvent, in which it is soluble. This problem has been encountered by Schiemenz and Thobe[142], and others. Secondly, in view of the very mild reaction conditions employed, we speculated on the possibility that the betaine intermediate (191), might be unusually stable. A lithium base had been used, and Schlosser and Christmann[143] have made an extensive study of betaine stability, in which they have shown that metal salts, especially those of lithium, form tightly associated ion pairs with betaines, which slow down their rate of decomposition. However, polar solvents and long reaction times break down these complexes.

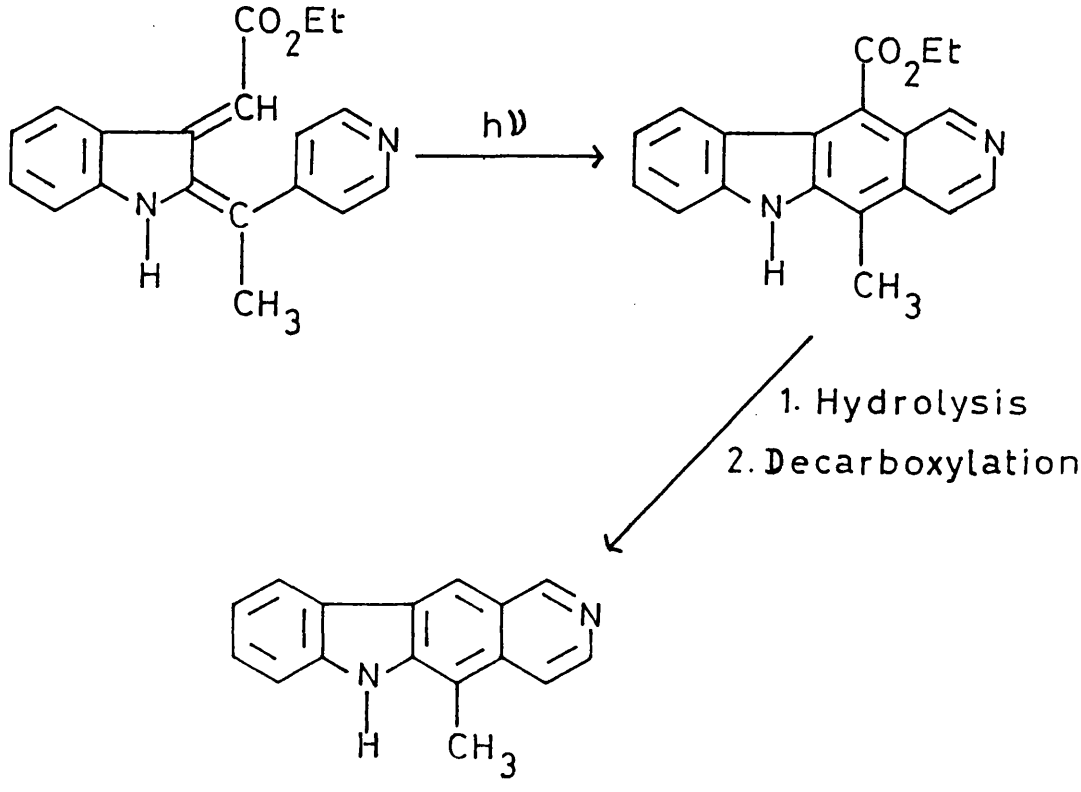
Accordingly, we dissolved the product in T.H.F. and heated it at reflux temperature for eight hours, but on examination of the residue a similar result was obtained. It was clear that triphenylphosphine oxide contamination of the product was occurring, so chromatography was attempted using both basic alumina and silica supports, and eluting with a variety of solvent systems (see experimental section p. 308), but despite much effort this met with no success. This problem proved to be intractable, because the indolinones are sufficiently polar, that most solvents capable of dissolving them also dissolve the phosphorus contaminant. House[144], comments that it is often difficult to separate the product from triphenylphosphine oxide in Wittig procedures when polar solvents have to be employed. In a bid to exploit the pyridine nitrogen atom as a means of forming a soluble salt of the product, dilute hydrochloric acid extraction was attempted, but this technique also failed to give a satisfactory result.

The often used alternative to the usual Wittig reaction in such cases is the Horner-Emmons modification[145], using triethylphosphonoacetate, which gives a water soluble phosphonate salt, that can be easily removed in the aqueous phase, as shown.



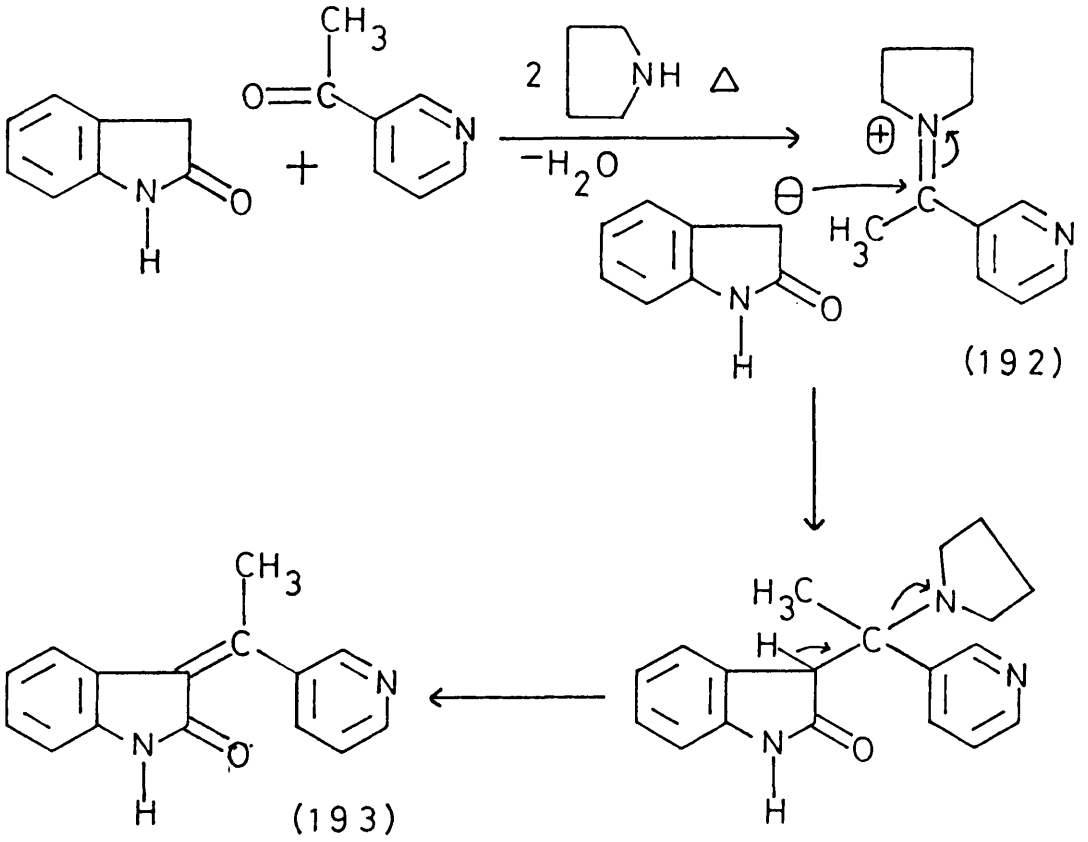
However, we did not make use of this alternative because the proposed reaction pathway outlined in (Scheme 62), would ultimately give rise to 11-desmethylellipticine, which has been shown to have greatly reduced biological activity[214] with respect to the parent alkaloid.

Scheme 62.



At this juncture we decided to discontinue the photochemical approach to ellipticine and turned our attention to a different method based on oxindoles. It has been known for some time in this laboratory, that 3-[1-(3-pyridyl)ethylidine]indolin-2-one (193) can be easily prepared as outlined in (Scheme 63).

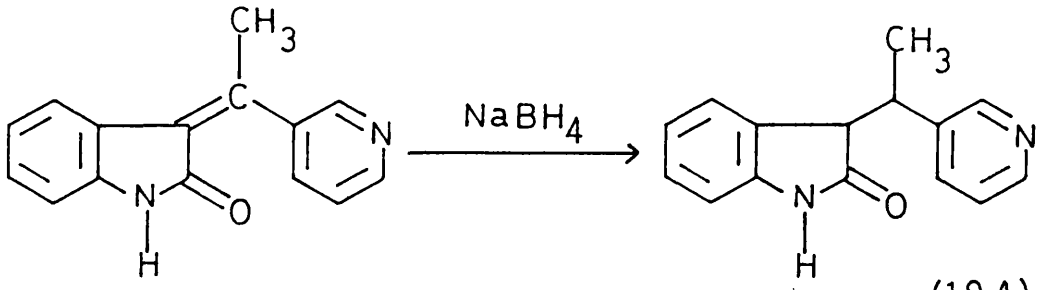
Scheme 63.



Oxindole and 3-acetylpyridine in the presence of pyrollidine give an 80% yield of the oxindolylidene (193) via the enamine system (192)[146] when Dean-Stark conditions are employed. One would expect both the E- and Z-forms of (193) to be produced in this reaction, but spectral data indicate that in fact, only the E-form is obtained[147,148] (see p171 for a discussion of the evidence

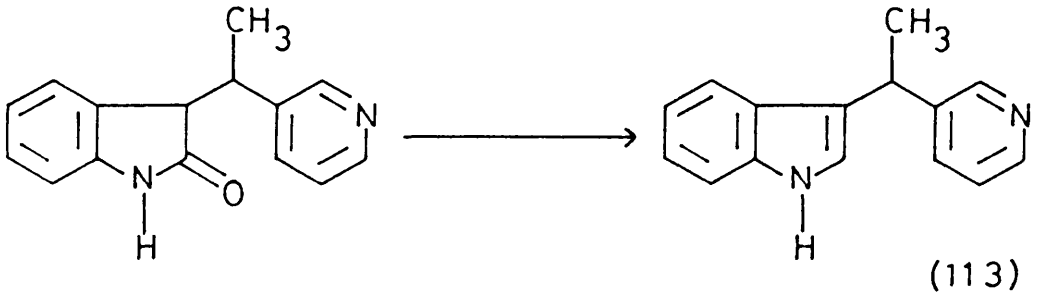
for this). However, for ease of presentation the molecule will be represented as the Z-form of (193).

Reduction of the oxindolylidene (193) with sodium boronhydride in ethanol at 60° affords the pyridyloxindole (194) in 90% yield.



(194)

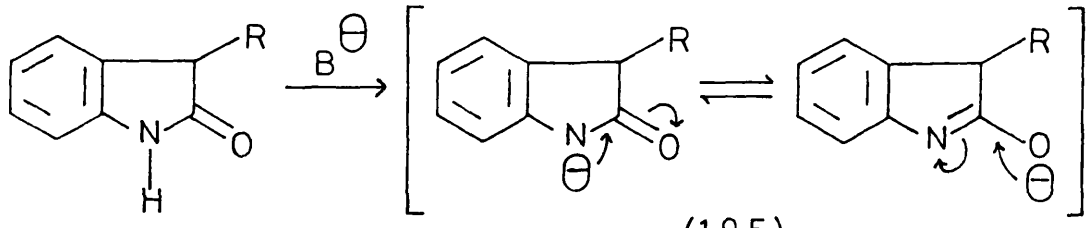
Subsequent reduction of the amidic carbonyl group would give rise to the indolylpyridylethane (113).



(113)

Unfortunately pyridyloxindoles of the type (194) have always proved to be inert to reduction. A number of groups have published work on the reduction of oxindole to indole systems. Lithium aluminium hydride L.A.H. effectively reduces N-alkylated oxindoles

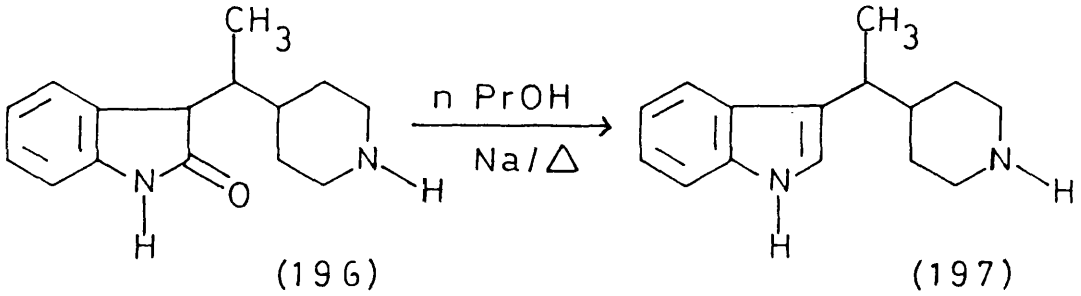
to indoles[149], but it is generally accepted that N-unsubstituted derivatives are not reduced by this reagent. This is thought to be due to the formation of the anion (195). This anion once formed, will resist further attack by nucleophilic species, thus innhibiting reduction of the carbonyl function.



(195)

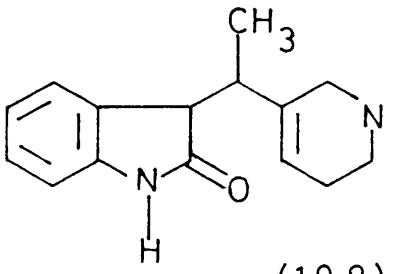
In support of this Kilminster[147] found that L.A.H. treatment of the pyridyloxindole (194) in boiling tetrahydrofuran gave only intractable tars.

The electron transfer reagent, sodium in alcohol has been used to reduce oxindoles, but once again the evidence suggests that only highly alkylated derivatives are converted to indoles[150]. The piperidyloxindole (196) has been reported to be reduced to the corresponding indole (197) by Italian workers[151] using sodium in boiling n-propanol.

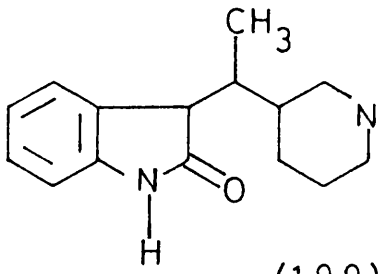


(196) (197)

Once again, however, experiments in this laboratory[147] showed that the pyridyloxindole (194) when treated in a similar manner gave only multicomponent mixtures, the mass spectra of which suggest the formation of the tetrahydro (198) and hexahydro (199) derivatives.



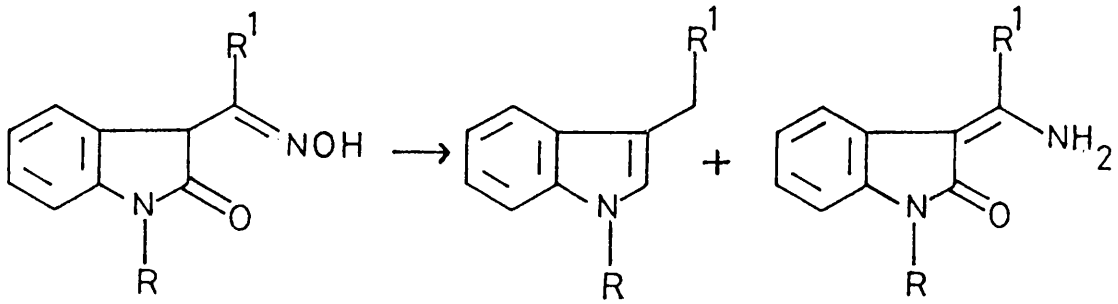
(198)



(199)

A disadvantage of this approach when applied to pyridyloxindoles, is that the electron transfer reagent will reduce the pyridine ring more readily than the amidic carbonyl group, and if successful the product would require dehydrogenation of the partially or fully reduced pyridine ring before completion of the remainder of the synthesis.

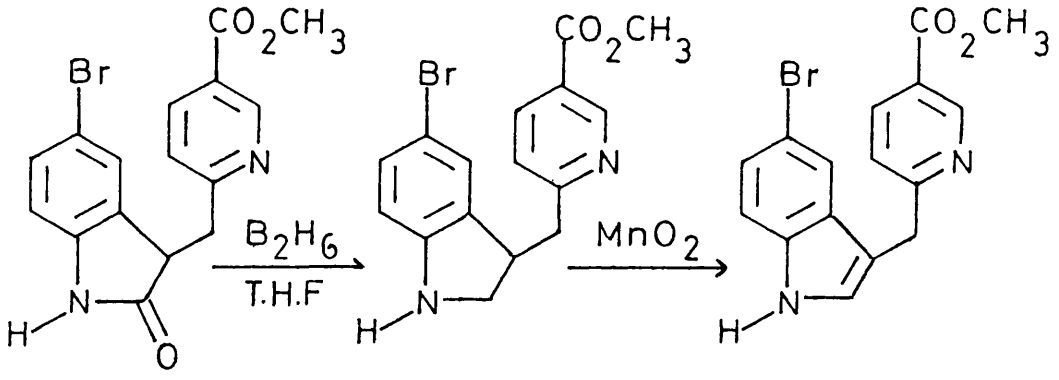
Wenkert[152] has catalytically hydrogenated 3-acyloxindole oximes (200) and obtained mixtures of 3-alkylindoles (201) and 3-(α-aminoalkylidine) oxindoles (202).



(200) (201) (202)

It is of interest to note, that here, N-unsubstituted compounds gave indolic products whereas N-alkyl derivatives gave only the ∝-aminoalkylidines.

Julia et al[153] successfully employed diborane to effect an indirect oxindole to indole conversion of lysergic acid analogues. Thus, the compound (203) was reduced to the indoline (204), which on treatment with manganese dioxide was oxidized to the indole derivative (205). Several other examples of the use of this reagent to reduce oxindoles have been reported.[154]

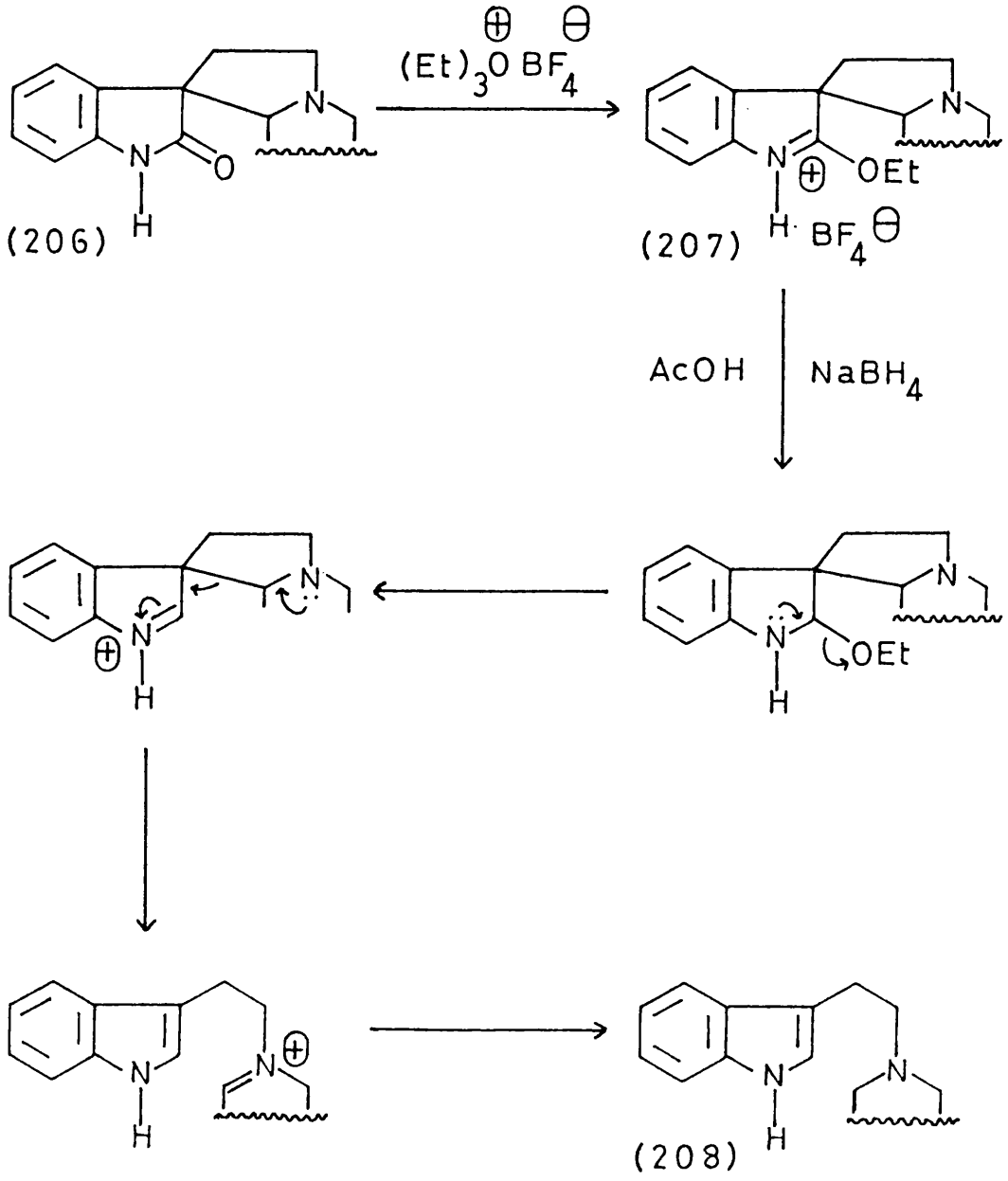


(203) (204) (205)

Despite the success of this approach with related compounds, work in this laboratory[147] shows that it is not effective when applied to the oxindole (194). Reactions using diborane under a variety of conditions were found to give starting materials only.

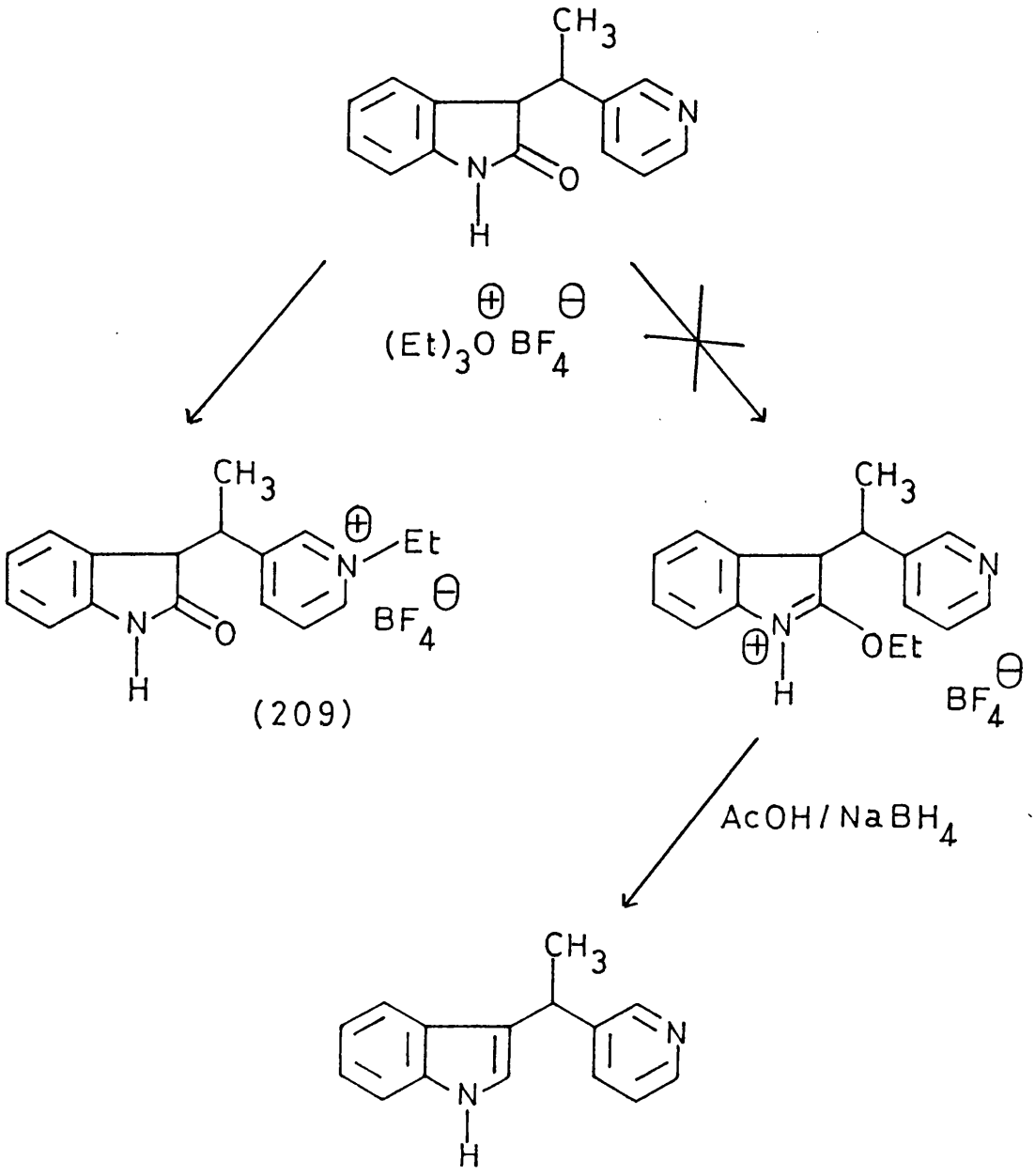
Aimi and co-workers[155] have reported the successful conversion of the oxindole ring of the alkaloid geissoshizine oxindole (206, only part structures are shown for ease of presentation), into the indole analogue (208) by treatment of (206) with triethyloxonium tetrafluoroborate to give the imine ether salt (207), followed by reduction of this with sodium borohydride in glacial acetic acid, presumably by the mechanism shown in (Scheme 64).

Scheme 64.



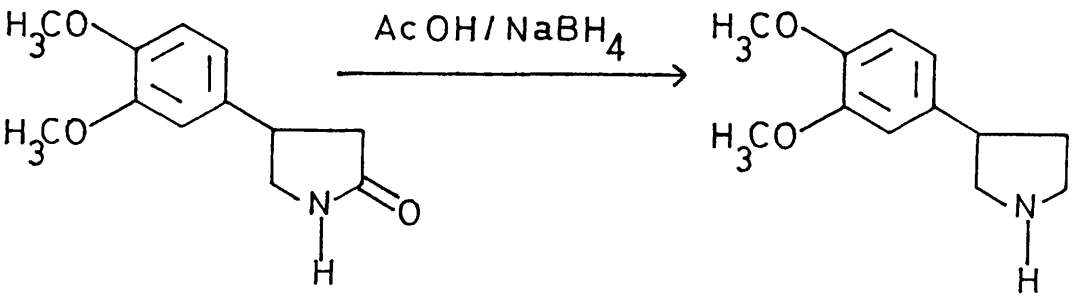
Attempts to apply this technique to the pyridyloxindole (194)[147], also failed and once again (see p.140), the sole product was the pyridinium salt (209) as outlined in (Scheme 65). In view of this result, the success observed with the alkaloid (206) seems rather surprising, as this compound also contains a basic nitrogen centre that might be expected to form a quaternary salt, but steric constraints in this more complex structure may prevent this.

Scheme 65.



Despite these earlier failures, the appeal of this general route to ellipticine precursors led to further attempts in this laboratory to effect the oxindole to indole conversion of (194).

Umino et al[156], have also used sodium acyloxyborohydrides for the reduction of carboxamides to amines. They report that primary and secondary amides are reduced by the use of an equimolar mixture of sodium borohydride and acetic acid. Thus, a 64% yield of N-ethylindoline was obtained from N-acetylindoline, and a 60% yield of the amine (211)from the amide (210).

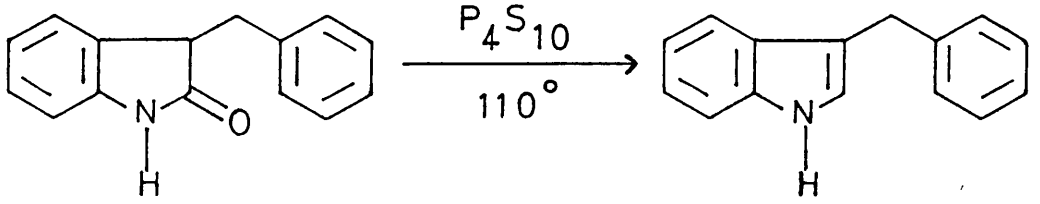


(210) (211)

Driver[148], investigated the application of this method to the oxindole (194), but reaction periods of up to twelve hours failed to reduce the carbonyl function, as did the use of trifluoroacetic acid in place of acetic acid, a technique that Umino claims will reduce tertiary amides.

Since N-alkylated oxindoles can be reduced to the corresponding indoles using L.A.H., the 6-alkyl ellipticines would be accessible by this method, but this line of enquiry was not pursued further because preliminary pharmacological investigations[223] indicate that these derivatives are not as biologically active as the 6H pyrido [4,3-b] carbazoles.

Plieninger and Werst[157] have reduced 3-benzyloxindole (212) to 3-benzylindole (213), using phosphorus pentasulphide (P_4S_{10}) at 110° for several hours, followed by treatment of the product with Raney nickel in ethanol.



(212) (213)

However, when applied to the oxindole (194)[148], this reduction procedure also failed.

Due to these discouraging results the oxindole approach to ellipticine was abandoned in this laboratory for some years, until we read with interest the recent report by Japanese workers Kubo and Nakai[158], who effected a number of oxindole to indole conversions as outlined in (Scheme 66).

Scheme 66.

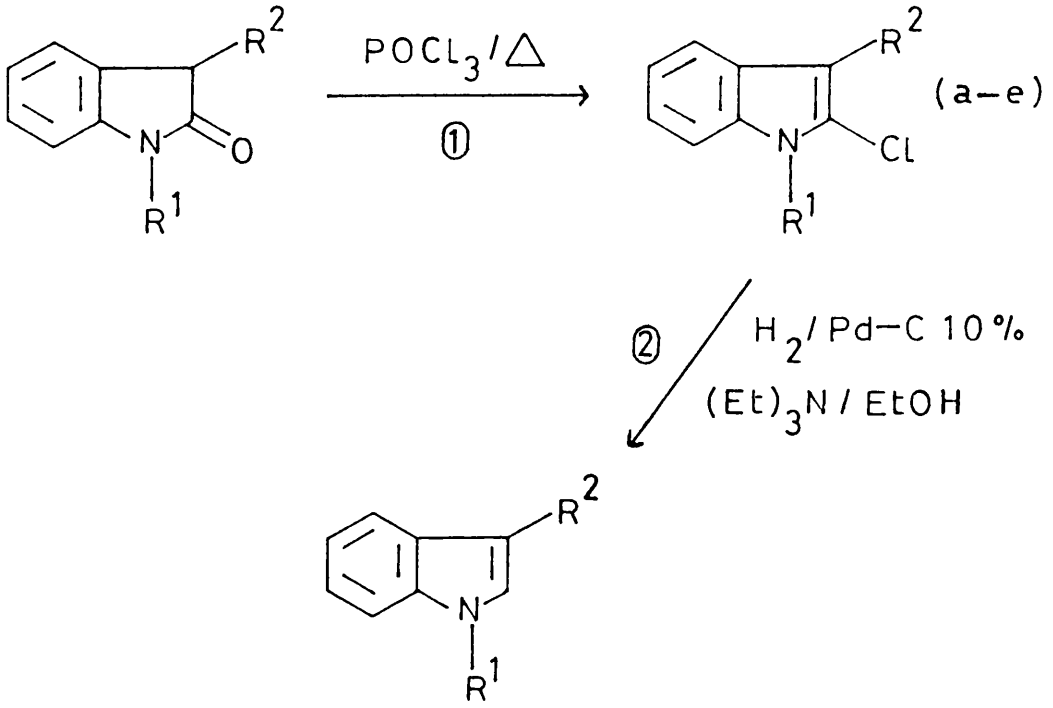
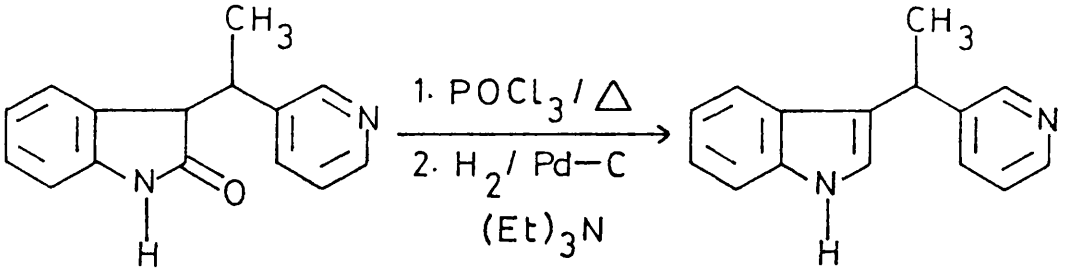


Table 3.

Product	R^1	R^2	Yield % (1)	Yield % (2)
a	H	CH_3	63	90
b	H	CH_2CH_3	93	89
c	H	$C_6H_5CH_2$	89	90
d	CH_3	C_2H_5	59	84
e	H	1-(3-pyridyl) ethyl	78	71

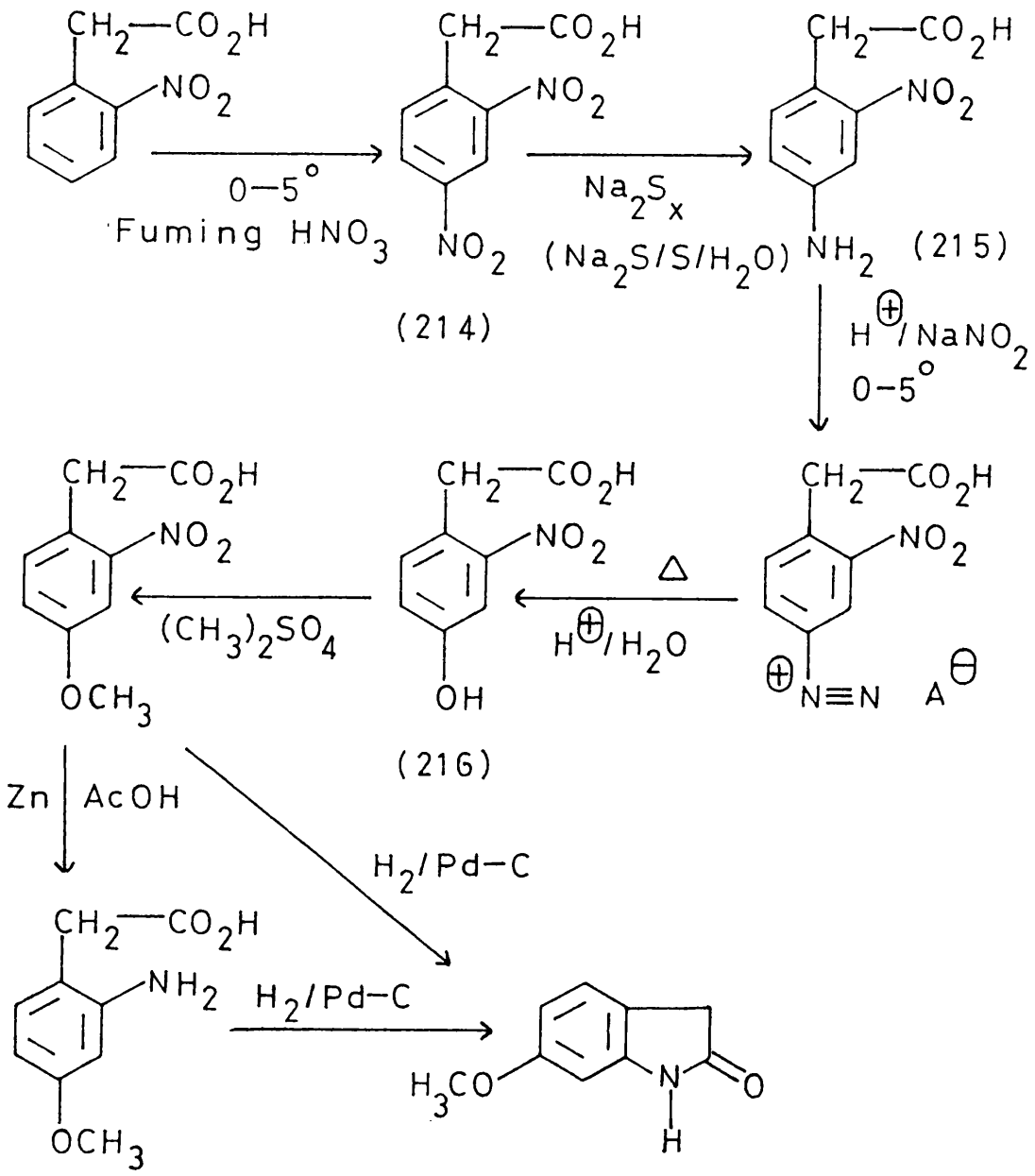
This appealingly short route involves heating the oxindole in neat phosphorus oxychloride under a nitrogen atmosphere to give the corresponding 2-chloroindole. The chloro group is then removed by catalytic hydrogenolysis using 10% palladium on carbon in the presence of approximately 2% of triethylamine to afford the desired indoles. Particularly interesting was the fact that one of these reductions (e in Table 3), involved the conversion of the oxindole (194) into the desired indole (113), which Kilminster[147] and Driver[148] had found so difficult to achieve with the methods available at this time.



(194) (113)

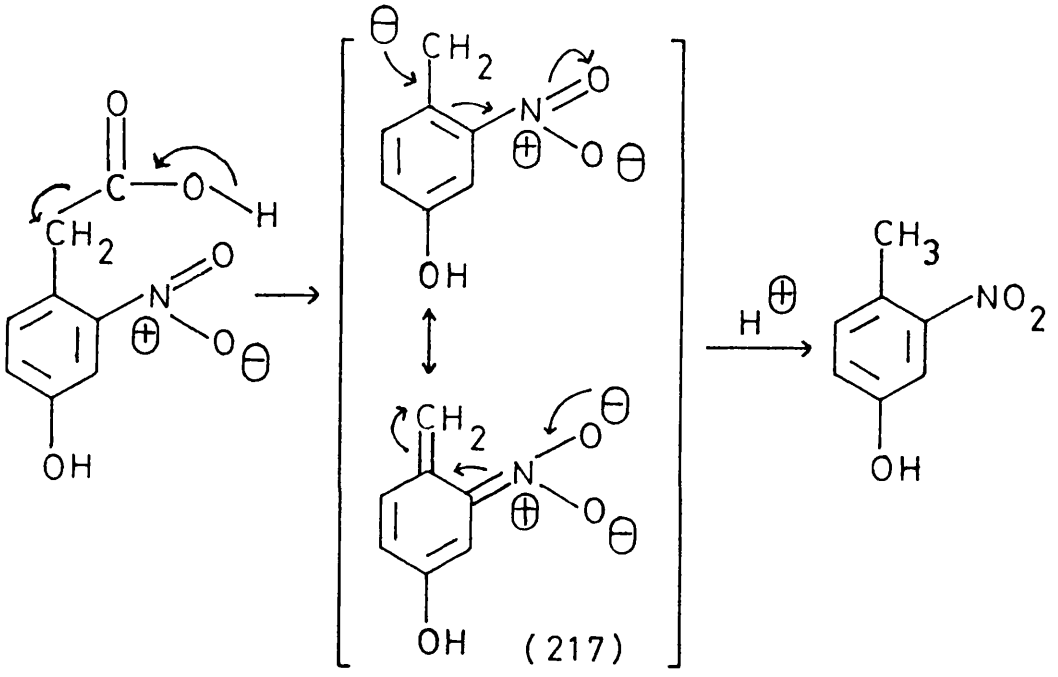
In view of this publication our interest in the oxindole approach to the synthesis of ellipticines revived, and we set about investigating the applicability of Kubo and Nakai's method to substrates that bore the desired 6-methoxyl function in the aryl ring of the oxindole moiety. In order to effect this aim it was necessary to prepare a quantity of 6-methoxyoxindole and our first attempt to synthesize it *via* 2-nitro-4-methoxyphenylacetic acid is outlined in (Scheme 67).

Scheme 67.



2-Nitrophenylacetic acid was nitrated[159] using fuming nitric acid at ice temperature to give 2,4-dinitrophenylacetic acid (214) as a white solid in 65% yield. The nitro group in the 4-position of 2,4-dinitrophenylacetic acid was selectively reduced using sodium polysulphide[104] to give 2-nitro-4-amino-phenylacetic acid (215) as orange plates from ethanol in 73% yield.

Diazotisation of (215) as the sulphuric acid salt[105], followed by treatment with hot aqueous acid, did not, however, give the desired 2-nitro-4-hydroxyphenylacetic acid (216), but 2-nitro-4-hydroxy-toluene(138) was the product The first formed acid (216) underwent spontaneous decarboxylation in the hot aqueous acid, as shown.



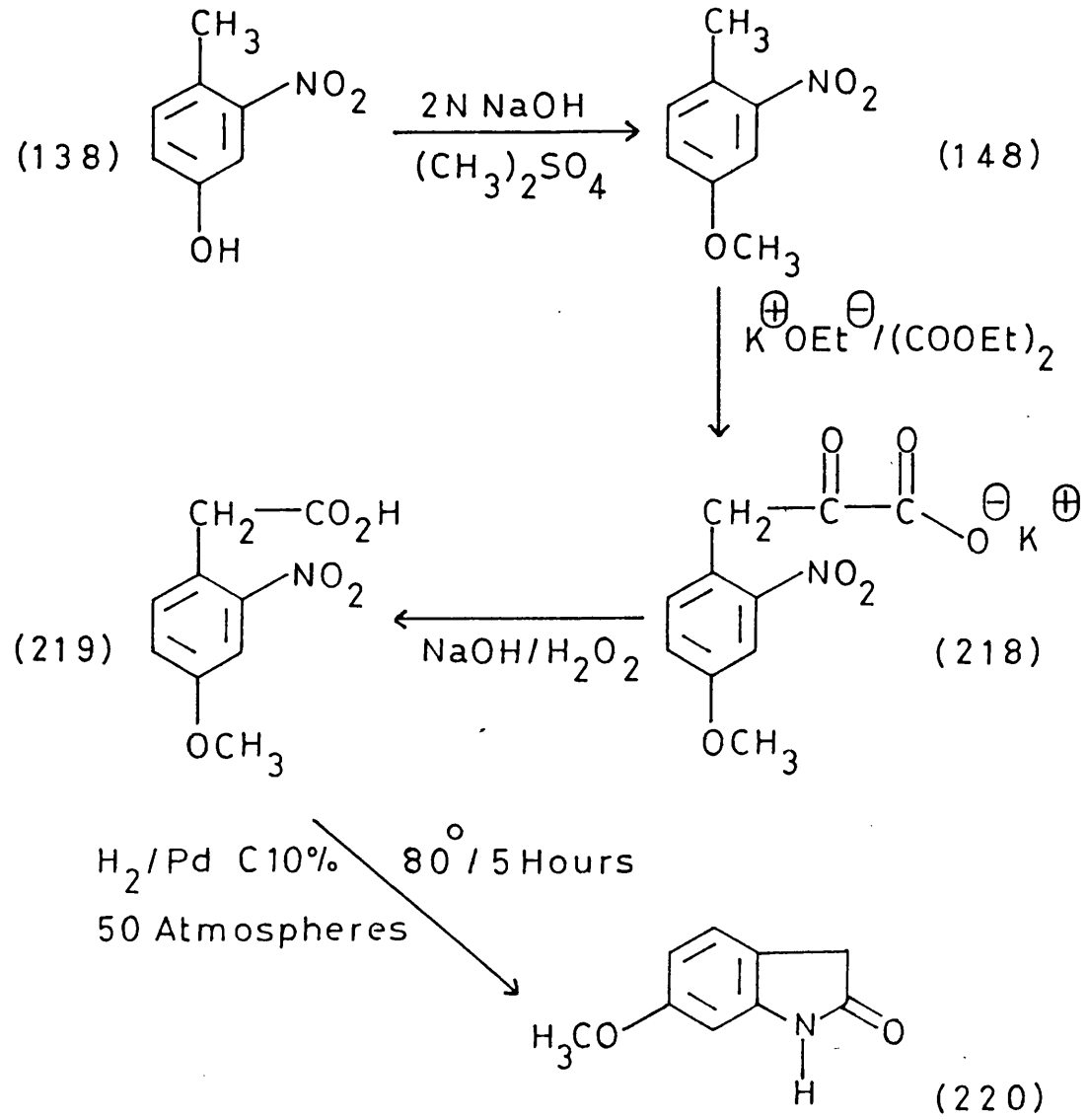
It might be expected that the anion (217) would experience a competitive effect between stabilisation by the 2-nitro group as shown and destabilisation by the 4-hydroxy function. But it would appear that the 2-nitro group remains the dominant influence as the reaction occurs more readily than with unsubstituted phenylacetic acid.[160]

Milder conditions in the phenol formation reaction using warm dilute hydrochloric acid rather than sulphuric acid did not improve the yield of the desired product (216). The

decarboxylation reaction occurred readily under the mildest conditions compatable with the conversion of the diazonium salt of (215) to the desired phenol. Thus, we were unable to implement the remaining stages of the synthesis shown in (Scheme 67).

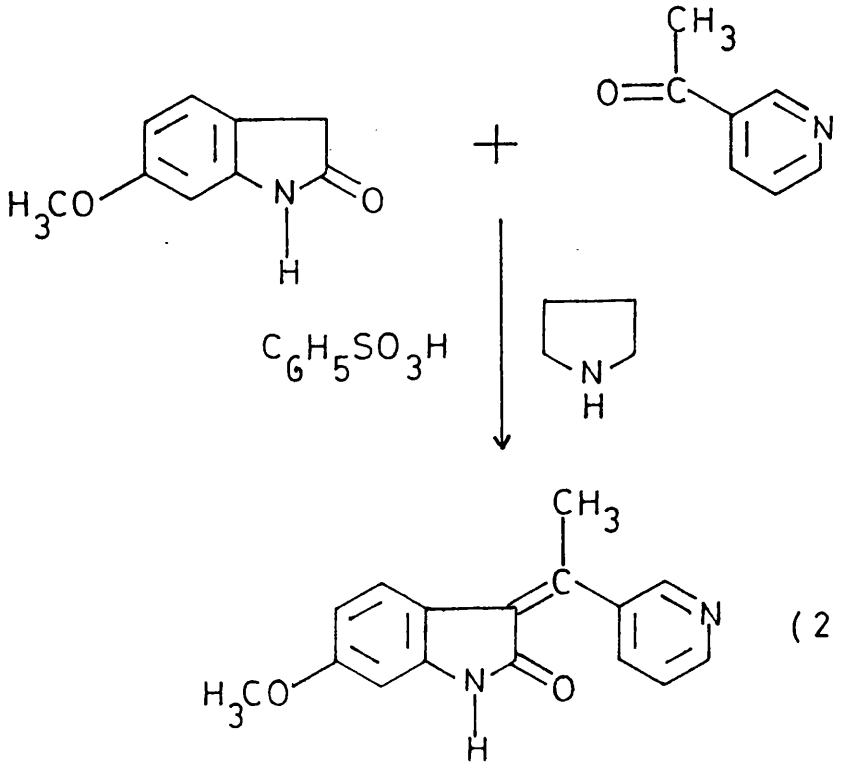
We now turned our attention to a more productive and somewhat shorter method of synthesis, favouring the route outlined in (Scheme 68). This employs 2-nitro-4-hydroxytoluene (138), obtained in the previous series of experiments, as a starting material, thereby redeeming some of the effort expended earlier.

Scheme 68.



2-Nitro-4-hydroxytoluene in dilute sodium hydroxide solution was methylated with dimethylsulphate to give 2-nitro-4-methoxytoluene (148) as a pale yellow liquid in 73% yield. This was condensed with diethyloxalate in the presence of potassium ethoxide to afford the pyruvate salt (218) as a bright orange solid. The salt (218) in sodium hydroxide solution was treated with 100 volume hydrogen peroxide[161], and on acidification gave 2-nitro-4-methoxyphenylacetic acid (219) in 68% yield. The acid (219) was dissolved in glacial acetic acid and ring closed under reductive conditions using 10% palladium on carbon under hydrogen in a high pressure autoclave at 80° for several hours.[162] This gave an 80% yield of pure 6-methoxyoxindole (220).

Having obtained our starting material we carried out the condensation with 3-acetylpyridine in the presence of pyrrolidine as before.



(221)

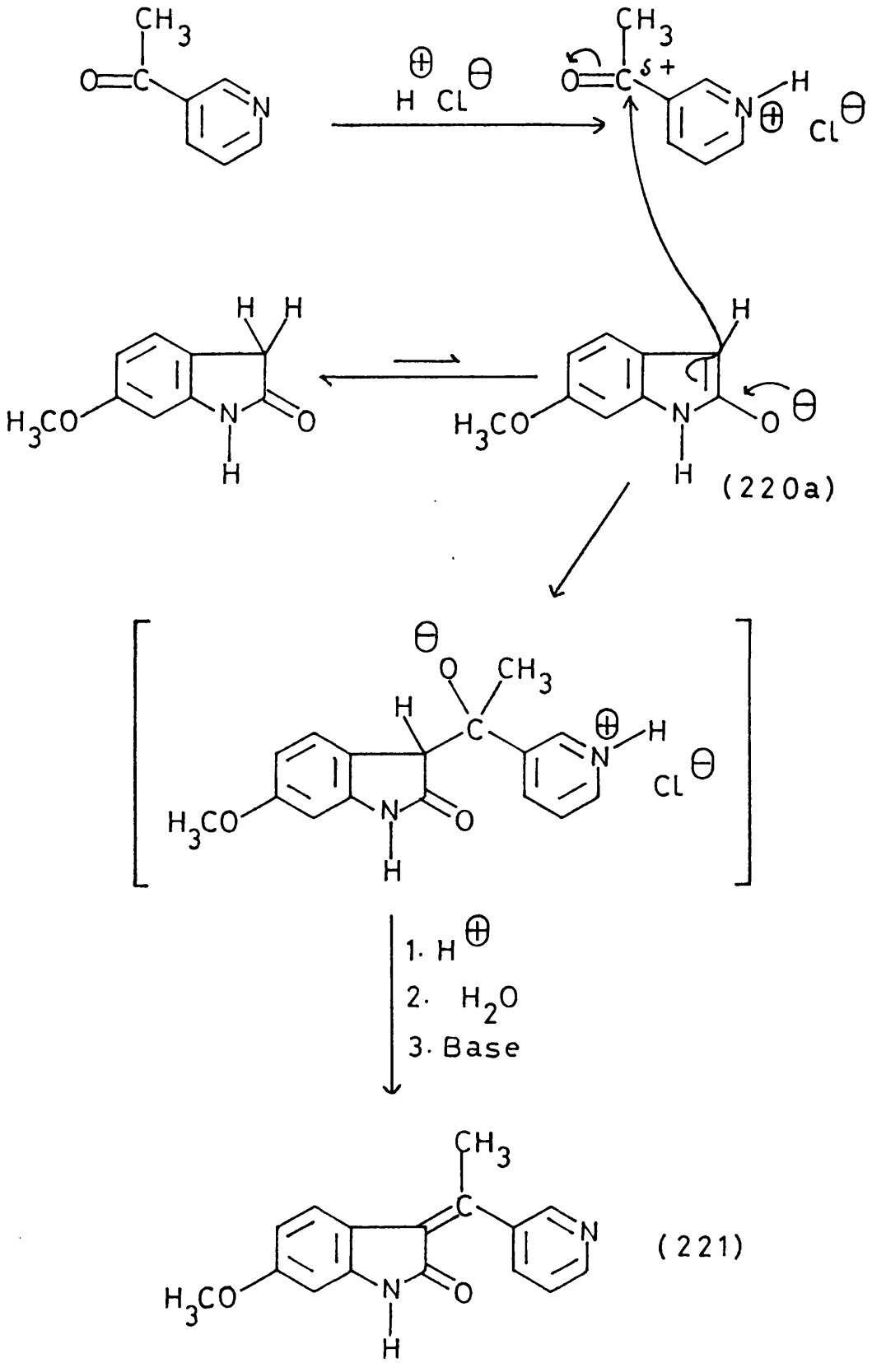
To our surprise on working up the reaction we could isolate none of the expected 6-methoxyoxindolylidene (221). The reaction was repeated using larger quantities of pyrrolidine and longer reaction times, but only dark coloured gums were obtained. Mass spectral analysis and T.L.C. confirmed that the only identifiable components were starting materials.

Since this reaction depends upon the formation of the C-3 oxindole anion, as shown in (Scheme 63, p.154), it would seem that the electron donating influence of the 6-methoxy substituent situated para- to this benzylic position, destabilizes its formation. Attempts employing stronger bases, including sodium hydride also failed to give any of the desired product These results lead us to speculate that the reaction might well proceed more readily under acid conditions.

6-Methoxyoxindole and 3-acetylpyridine were heated together in methanol saturated with dry hydrogen chloride gas, at 180° for eight hours. This treatment gave a bright orange salt, that on basification with sodium hydrogen carbonate solution afforded a canary yellow solid. This did indeed prove to be the required 6-methoxyoxindolylidene (221) in 70% yield. This presumably occurs via the mechanism shown in (Scheme 69). The first formed 3-acetylpyridinium chloride may react with the oxindole through a low equilibrium concentration of the tautomer (220a)[163] Daisly[164], Beckett and Walker, indicate that a tendency to form trace amounts of such tautomers is more likely with electron rich systems such as the methoxy derivatives. Protonation, elimination of water and

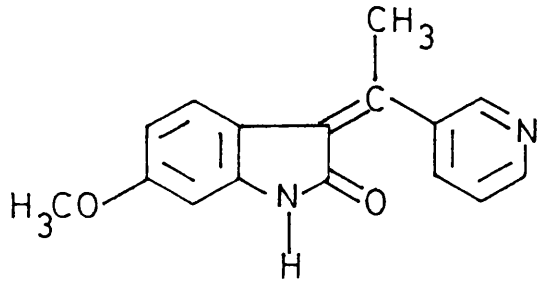
basification then give the 6-methoxyoxindolylidene (221).

Scheme 69

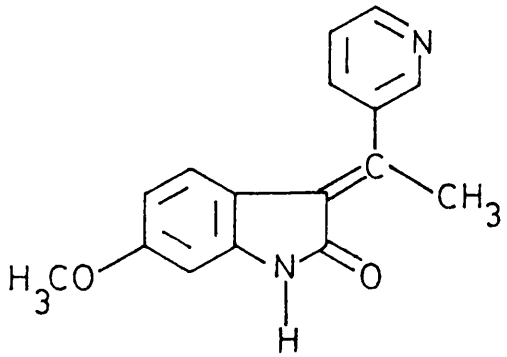


The infrared spectrum of (221) displayed peaks at ν_{max} 3150, 1685 and 1610cm^{-1}, corresponding to the N-H group, the carbonyl function and the carbon-carbon double bond, exocyclic to the oxindole ring, respectively. Mass spectrometry indicated the expected molecular ion at a mass to charge ratio of (m/e$^{+\cdot}$ 266), and a peak at (m/e 251) due to loss of a methyl group, which is characteristic of these compounds. The 100 MHz ^{1}H n.m.r. spectrum is in agreement with what one would expect for this structure and is reproduced in (Figure 5).

It is evident from the spectral data that the condensation reaction furnished only one of the two possible isomers shown.



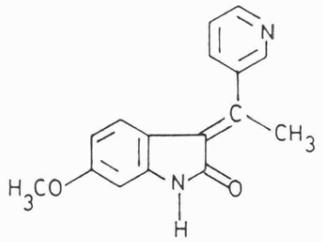
(Z 221)



(E 222)

If both geometrical isomers E- and Z- were present in the product one would expect to observe two methyl resonances at different chemical shift positions. The methyl group in the E-isomer would occur at lower field with respect to that from the Z-form, because the former lies in the deshielding zone of the carbonyl

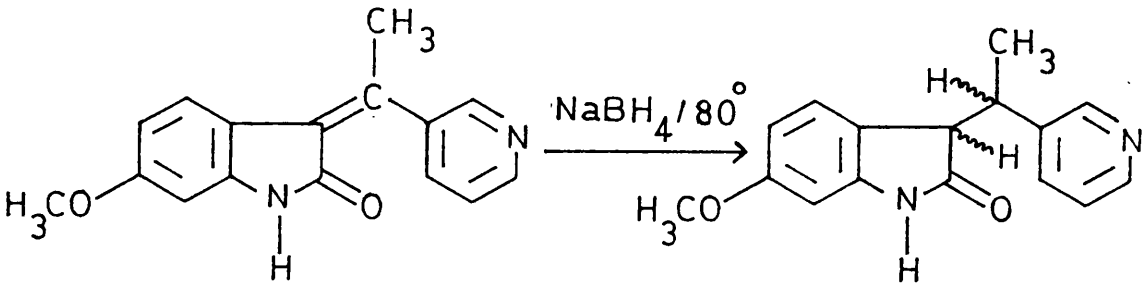
(Figure 5)



(222 E)

function. The observed value of 2.60 p.p.m. suggests that the isomer obtained is in fact the E-form (222). However, for ease of presentation the molecule is represented as (221Z).

The 6-methoxyoxindolylidine (221) was reduced with sodium borohydride in ethanol[147] at 80° to give the oxindole (223) as a diastereomeric mixture.

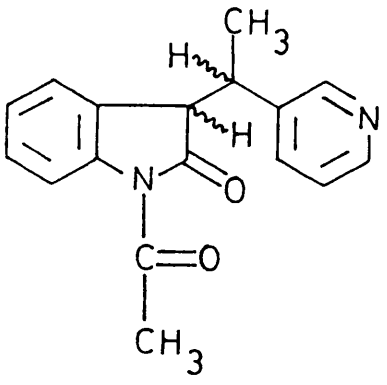


(223)

The product oil proved very difficult to crystallize, but prolonged trituration with aqueous ethanol gave a small quantity of a white solid. The mass spectra of the oil and the solid were identical giving a molecular ion at m/e$^{+\cdot}$ 268 corresponding to the reduced compound (223). The infrared spectrum exhibited two carbonyl absorptions at ν_{max} 1700 and 1750 cm^{-1} and the 100 MHz ^{1}H n.m.r. spectrum also displayed two sets of signals suggesting that the reduction product (223) is a mixture of diastereoisomers.

These findings are in complete accordance with those of Kilminster[147], who prepared the N-acetyl compound (224) at an earlier time in this laboratory. This compound is easier to crystallize

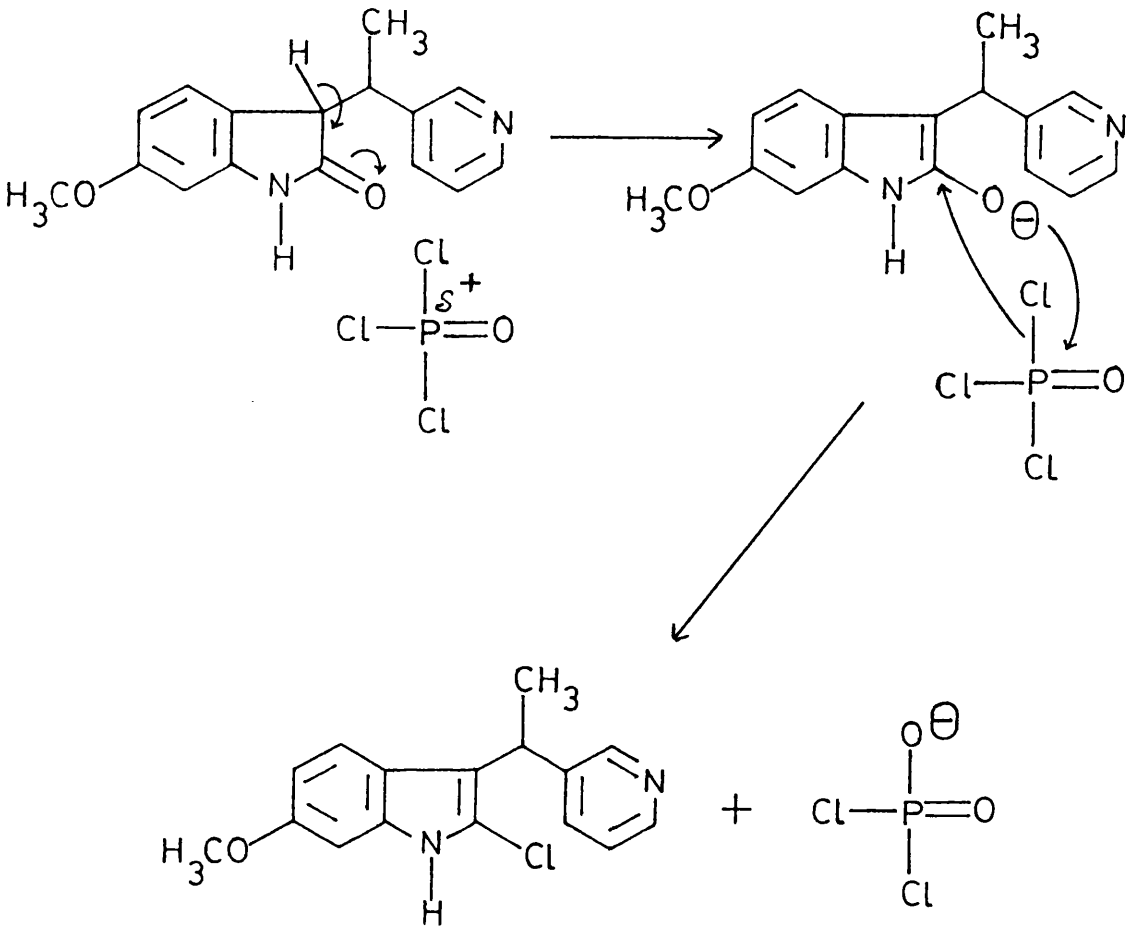
than the parent molecule and the ^{1}H n.m.r spectrum of (224) proved beyond doubt that compounds of this type exist as diastereoisomers.



(224)

The bridge head methyl group was shown as two equal sized three proton doublets, with a chemical shift difference of 0.25 p.p.m. The N-acetyl methyl function was represented by two, three proton singlets separated by 0.15 p.p.m.

Satisfied that our reduced product was simply a mixture of diastereoisomers we did not attempt to crystallize the bulk of the product because earlier work with the unsubstituted compound indicated that this would be very time consuming and tedious. We therefore carried the product oil forward to the next stage of the synthesis, which involves attack of phosphorus oxychloride on oxygen of the carbonyl function to give the chloroindole (225).

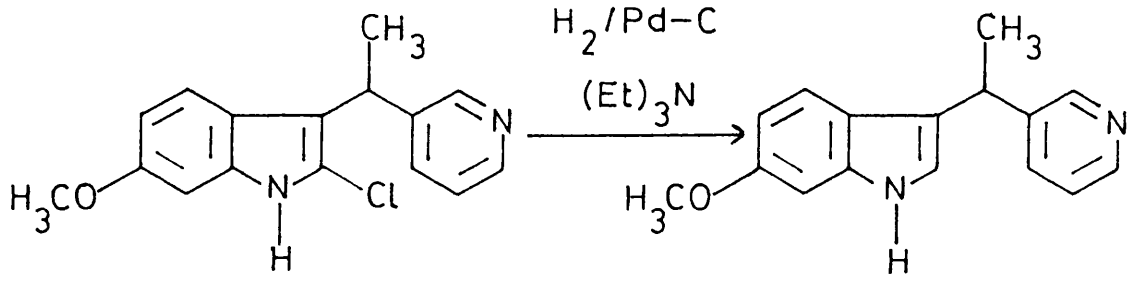


(225)

A mixture of the 6-methoxyoxindole (223) and phosphorus oxychloride were heated together at 110° under a nitrogen atmosphere for five hours. After a base work up procedure (see experimental section p. 321), the product was obtained from ethyl acetate as a green coloured gum. This was purified by chromatography on silica gel, eluting with ethyl acetate which contained a small amount of methanol. The pure compound was then obtained by crystallization from ethanol.

The mass spectrum shows a molecular-ion isotope cluster typical of a chlorinated aromatic compound with m/e (% relative intensity) values at 289 (M + 2, 20), and 287 ($M^{+\cdot}$, 63). The infared spectrum does not display a carbonyl absorption, but has a peak at ν_{max} 745cm^{-1} indicating that a carbon-chlorine bond is present. The ultraviolet spectrum exhibits bands normally associated with indole absorption and the 100MHz ^{1}H n.m.r. spectrum recorded in d^{6} dimethylsulphoxide displays the following resonances in p.p.m. A three proton doublet at 1.80 representing the bridge head methyl group, a second three proton singlet at 3.82 for the methoxyl group protons, and the lone proton on the bridge head as a quartet at 4.50. Seven aromatic signals occur between 6.0 and 8.50 and the lone N-H proton at 11.60. Satisfied that the physical data were entirely consistent with the structure (225) we continued to the next stage of the synthesis.

Hydrogenolysis of the 6-methoxychlorindole (225) to give the desired compound (161) proved much more difficult than reported by Kubo and Nakai for the parent compound.



(225) (161)

Initially we followed the published procedure, and the chloroindole (225) in ethanol, together with an equal weight of 10% palladium on carbon was hydrogenated in the presence of 2% triethylamine at room temperature. It is significant that Kubo and Nakai do not quote the hydrogen pressure they employed, but for our first reaction we used approximately three atmospheres by analogy with related reactions[165,166]. However, after four hours, T.L.C. and mass spectral analysis showed that only starting material was present. It was felt that these reaction conditions were not sufficiently energetic so we modified them as follows.

Firstly, the temperature was increased to 60° and the time taken for the reaction prolonged to seven hours. On working up this reaction, T.L.C. and mass spectral evidence indicated that some of the desired product (161) had formed, but it was mixed with a considerable amount of unreacted starting material. The results of this experiment clearly show that very vigorous conditions are required for success with this reaction, thus we carried out a high pressure experiment.

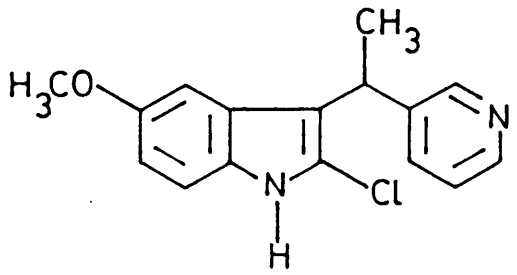
The hydrogenolysis was carried out using a pressure of seventeen atmospheres at 60° for sixteen hours. On working up this reaction it was found that almost all of the chloroindole (225) had been converted into the product (161). Traces of starting material were removed by chromatography and crystallization to give a pure product in 56% yield.

We felt that the difficulties experienced with this reaction might be due to catalyst poisons, but repeating the reaction with a fresh sample, did nothing to improve the results. Horner, Reuter and Herraman[167] quote amines as being catalyst poisons under certain conditions, so we carried out the reaction in the absence of triethylamine, but in this case no reaction occurred.

The use of Adams platinum oxide catalyst, higher pressures and temperatures in excess of those previously employed were discounted because of the possibility of further reduction to a mixture of indoline structures. Adkins and Burks[168] have studied the catalytic reduction of indole and comment that such techniques always reduce the indole ring system to a mixture of products dependent on the temperature, pressure and catalyst employed. The 3-substituted derivatives such as (225) are indeed harder to reduce[169] than unsubstituted indole. But we felt that such a reaction would require much time consuming experimentation to achieve an optimum yield of product without undesirable side reactions.

We concluded that the slower reaction observed must, once again, be due to the 6-methoxyl substituent. This may be rationalised as a consequence of the increased electron availability in the indole nucleus. This will compete with the inductive effect of the chlorine atom, resulting in a less polarised carbon-chlorine bond. Such a bond would be less easy to cleave and may result in a slower reaction.

If this rationalization is correct it might be expected that the 5-methoxy isomer (226) would present an even more unfavourable substrate for hydrogenolysis, due to the electron releasing para-effect on the adjacent nitrogen of the indole ring.



(226)

Further work in this laboratory by other members of the group has confirmed this. The 5-methoxy compound (226) was found to be almost inert to hydrogenolysis unless such extreme conditions were used as to risk further reduction and degradation of the product.

At this point we decided to carry out a reappraisal of our earlier work concerning the Grignard condensation of 6-methoxyindole with 3-(1-chloroethyl)pyridine (see p. 117). The high proportion of unreacted 6-methoxyindole recovered from these reactions caused us to consider the possibility that the 6-methoxyindolylmagnesium bromide might not be reacting as required. Tests on this intermediate showed it to have an unexpectedly low solubility in ether, and to be present in the reaction mixture as a very fine suspension rather than a solution. This fact was clearly

contributing to the lower yields obtained, and the following modifications were carried out.

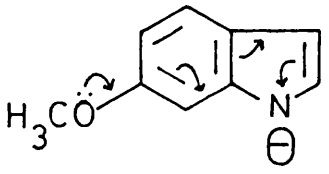
The experiment was conducted as previously described on (p. 118), but just sufficient dry tetrahydrofuran was added to dissolve the suspension of 6-methoxyindolylmagnesium bromide and form a clear homogeneous solution. After removal of the T.H.F., followed by the usual work-up procedure (see experimental section p. 281), the indolylpyridylethane (161) was obtained in 40% yield.

Encouraged by the result of this experiment we next carried out the reaction using dry T.H.F. as the only solvent. This gave a 43% yield of a brown and rather impure product that required treatment with charcoal, chromatography and crystallization before it was acceptably pure, but the yield of pure material was only 28%. The impurity of the original products from the reactions carried out in pure T.H.F. appear to be due to a proportion of included polymeric material. This presumably arises because of the much greater solvating power of T.H.F. with respect to ether.

Finally we concluded that the optimum yield for this method is in the region of 40% and can be obtained by using a mixture of diethylether and tetrahydrofuran in a ratio of 4:1 at room temperature. We considered this result to represent a more attractive synthetic pathway than the modification of Kubo and Nakai's method, because the Grignard condensation gives the desired product in a single step, and we now used this reaction to prepare a sufficiently large quantity of the indolylpyridylethane (161), to continue the synthesis of 8-hydroxyellipticine.

It is interesting to note in passing that, contrary to expectations, this reaction is slightly less favourable with a 6-methoxy substituent than with indole itself.[53,54]

At present we are unable to give a precise rationalization of these effects, but it is probable that the 6-methoxy substituent tends to innibit formation of the appropriate indolylmagnesium bromide by its ability to destabilize the anion (227) with respect to that from indole.

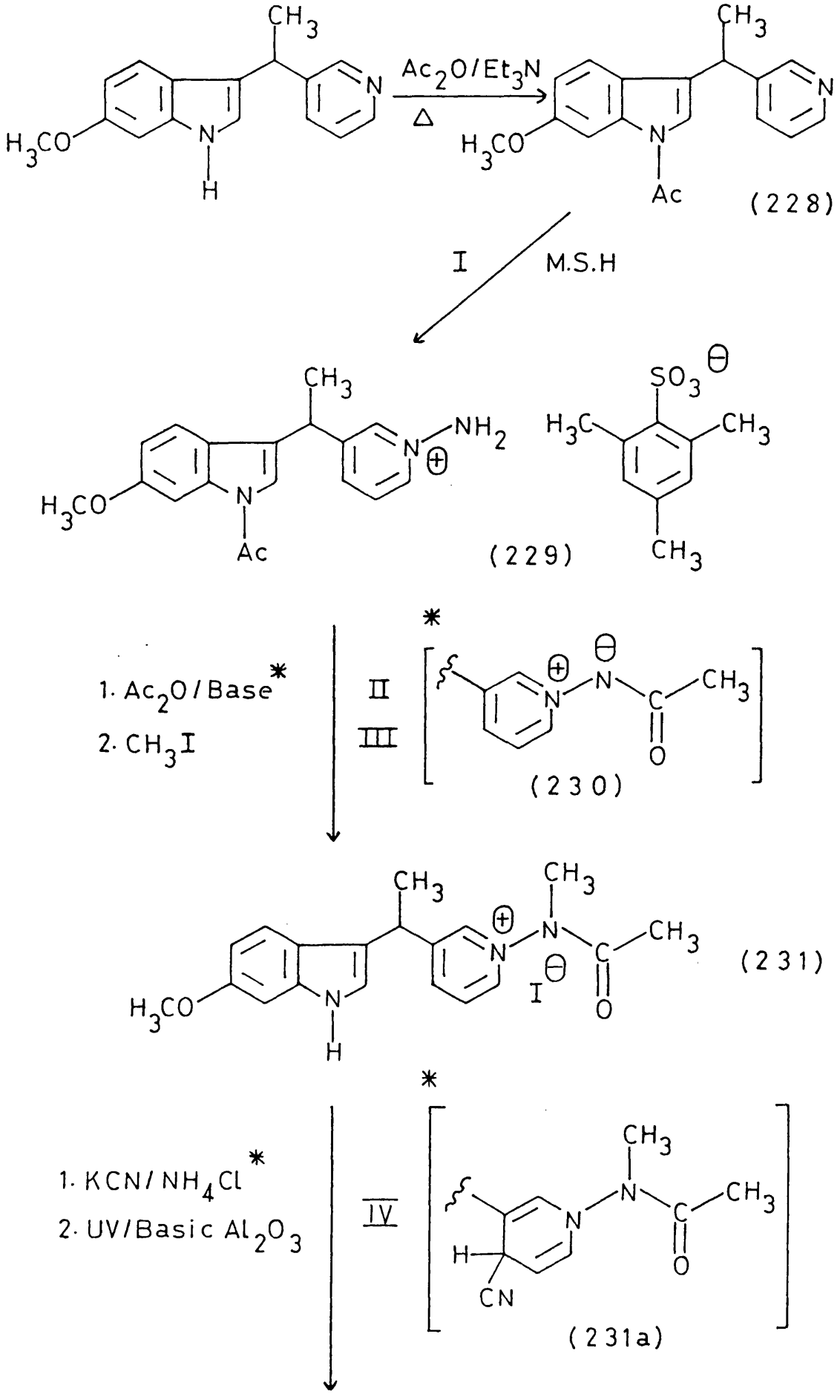


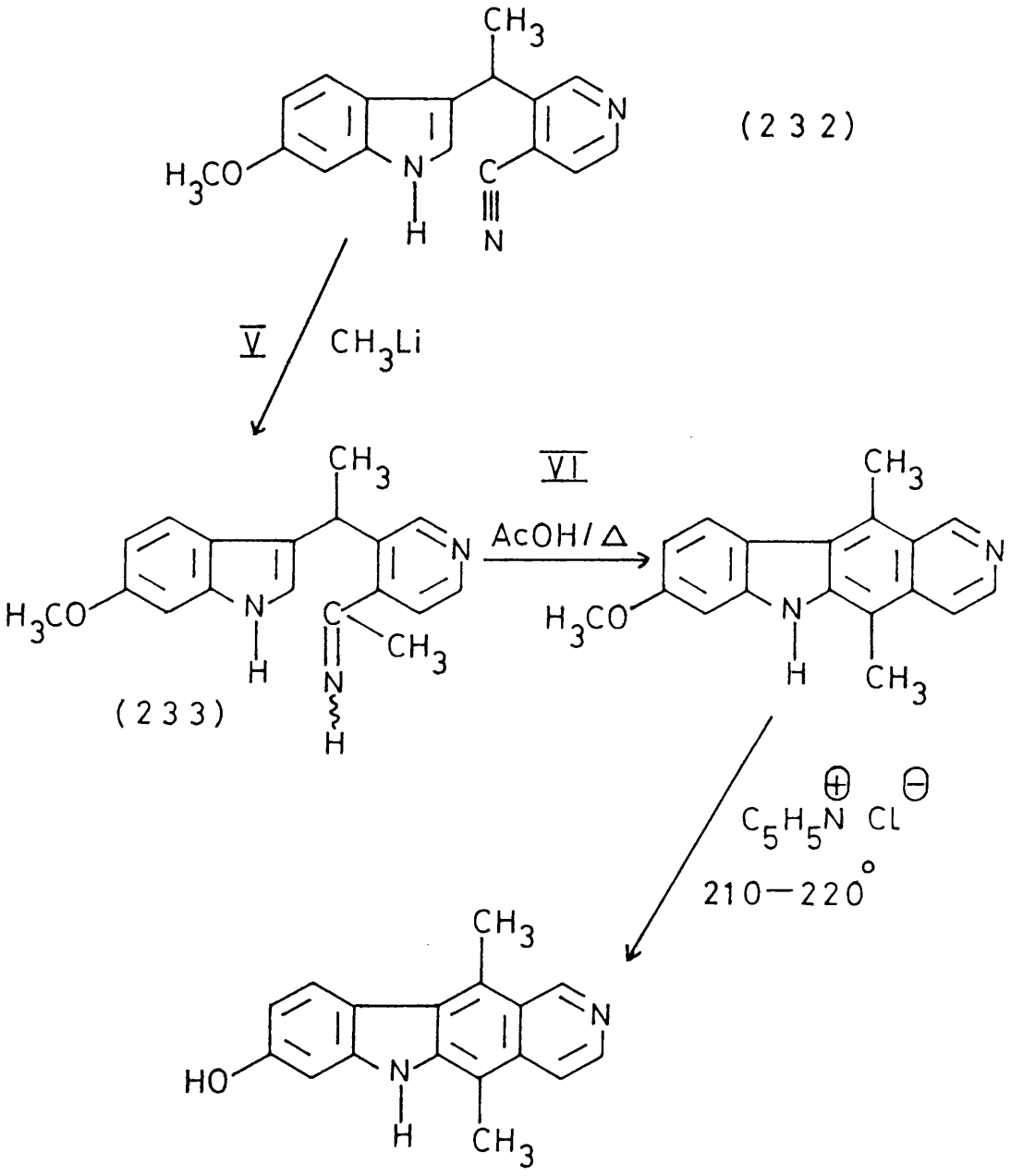
(227)

The considerable amount of unreacted 6-methoxyindole isolated from all the above experiments would tend to support this conclusion. It can also be argued that the case of 5-methoxyindole would give rise to even poorer yields in this reaction, due to the ability of this compound to exercise a <u>para</u> effect and further work in this laboratory supports this contention. Yields of 30% or less of the appropriate indolylpyridylethane are obtained when 5-methoxyindole is used as the substrate.

Having obtained the indolylpyridylethane (161), the remaining stages of the synthesis were implemented as shown in (Scheme 70).

Scheme 70


Ac2O/Et3N
Δ
(228)
I
M.S.H
(229)
1. Ac2O/Base*
2. CH3I
II
III
(230)
1. KCN/NH4Cl*
2. UV/Basic Al2O3
IV
(231)
(231a)



It proved necessary to acylate the ring 'B' nitrogen atom of the indolylpyridylethane (161) prior to amination of the pyridine nitrogen, and this was achieved using acetic annhydride in the presence of triethylamine. Thirty minutes at reflux temperature afforded an 87% yield of the acetyl derivative (228). Treatment of (228) with mesitylene sulphonylhydroxylamine (M S.H.) afforded the N-amino salt (229), which was acetylated with acetic annhydride to give the zwitter ion (230). Methylation with methyl

iodide furnished the methiodide (231), which was dissolved in water and treated with potassium cyanide in the presence of ammonium chloride to yield the 1,4-dihydrocyanopyridine intermediate (231a) which was not isolated, but directly exposed to ultraviolet light. The product was passed through a short column of basic alumina, eluting with a mixture of chloroform and methanol in a ratio of 98:2, this furnished the pure carbonitrile (232). Attack upon the cyano function by methyl lithium gave the imine (233). This was hydrolysed using 20% aqueous acetic acid at 80° for fifteen minutes, which effected both ring closure and aromatization to give 8-methoxyellipticine in 42% overall yield from the indolylpyridylethane (161).

O-Demethylation of the 8-methoxyl function was achieved by application of the published procedure employed by Dat-Xuong et al[62]. 8-Methoxyellipticine was heated with pyridinium chloride at 210-220° for thirty five minutes, to give a 30% yield of 8-hydroxyellipticine.

This route combines the advantages of a one step condensation with mild conditions in all stages prior to, and including the final ring closure. This is particularly valuable as experience has led us to regard some of the intermediates en route to the ellipticine ring system as thermally unstable. The moderate yield of the final O-demethylation step cannot be due to instability of the ellipticine ring system as many of these compounds can be sublimed unchanged. However, the hydroxy function may well induce some

oxidative degradation at the high temperatures required. Attempts to reduce the severity of the demethylation conditions did not improve the yield, and T.L.C. showed the reaction to be incomplete if lower temperatures or shorter reaction periods were employed.

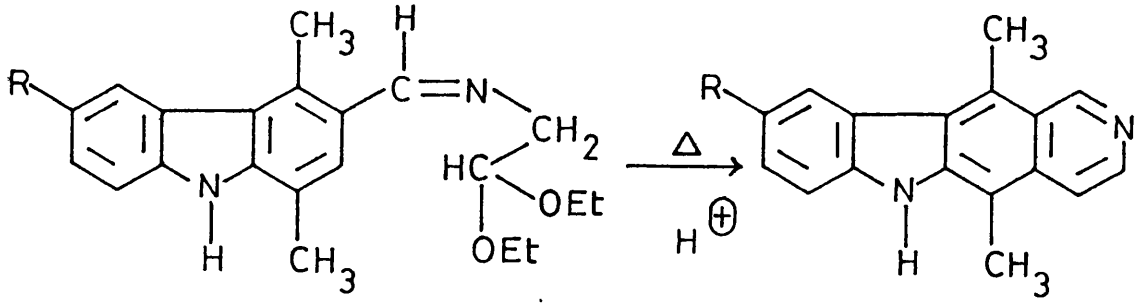
This work provides the first synthesis of 8-hydroxyellipticine (Tetrahedron Letters; 1981, 22, 2119) and (J.C.S. Perkin I; 1982, 2, 587), which confirms the structural assignment of Rosazza and Chein[63], who, as previously stated, isolated this compound as an ellipticine metabolite of the micro organism Aspergillus aliaceus (p. 67). Limited time precluded the use of this method for large scale preparation, however, it is hoped that this may be carried out in the future to allow detailed pharmacological evaluation.

Having successfully completed the synthesis of 8-hydroxyellipticine we turned our attention to a study of the direct electrophilic substitution of the ellipticine ring system. This approach has been little investigated, and in the past most workers have built in the required substitution by means of quite lengthy routes, often giving rather low overall yields (see introductory section).

The lack of interest in direct substitution has been due to the harsh conditions usually employed, which tend to degrade the alkaloid, and the possibility of ambiguity in the position of substitution. We have now established mild conditions for the introduction of two different substituents and defined the position of substitution in the 'A' ring, in each case by means of a high resolution ^{1}H n.m.r. examination and comparison with other physical data obtained from authentic samples, prepared by more lengthy conventional methods.

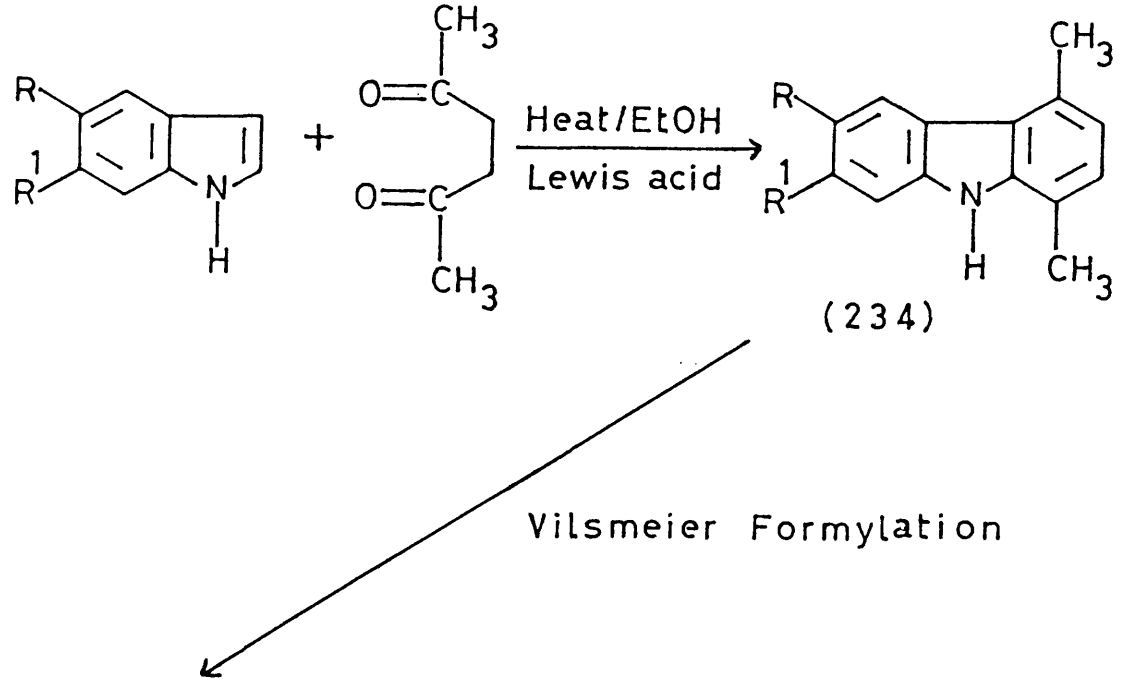
In order to implement this study we required a stock of ellipticine, and we chose to adapt the method of synthesis used by Dalton[12] as this is generally the shortest and most satisfactory approach to the unsubstituted molecule However, we felt that the yields at several stages could be improved by the use of milder conditions.

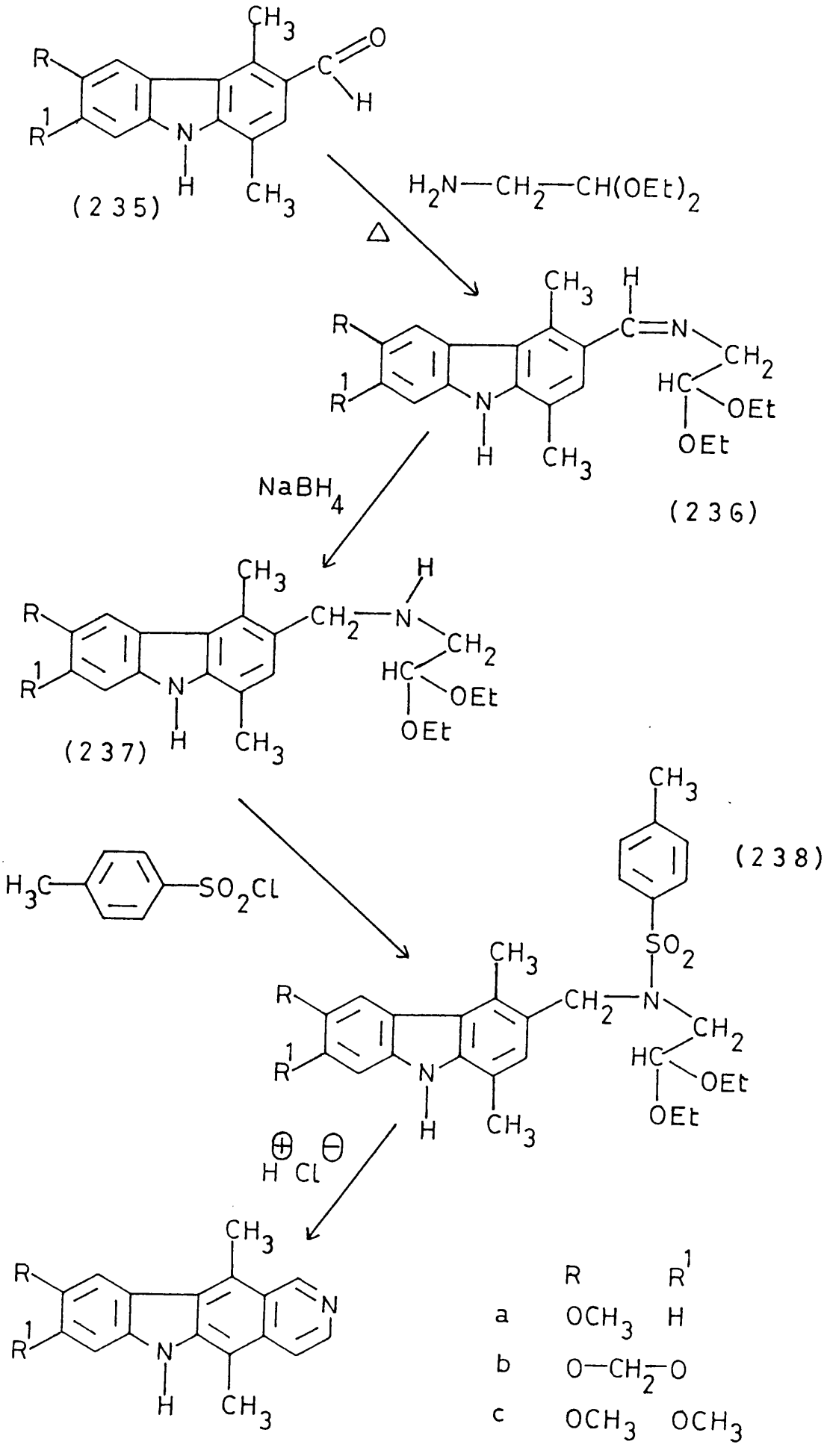
One of the main problems of the general route used by Dalton (see p. 11), is the severity of the conditions used in the final Pomeranz-Fritsch isoquinoline ring closure.



Originally this reaction was conducted at 150° in 100% ortho-phosphoric acid, but these are harsh conditions and undoubtedly cause a certain amount of breakdown to occur. In order to overcome this problem, Guthrie and his team[170] in the U.S.A. used the milder modification outlined in (Scheme 71), to prepare a variety of 9-alkoxy substituted ellipticines.

Scheme 71

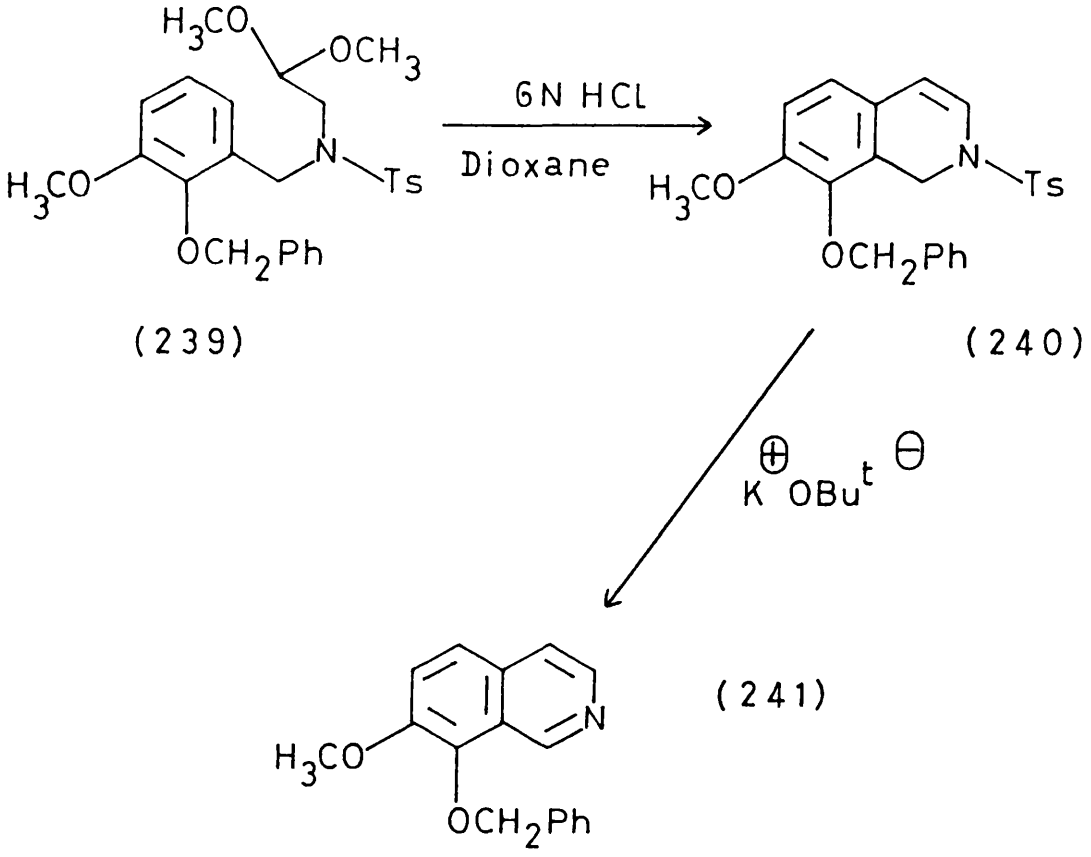



(235)
(236)
(237)
(238)
NaBH4
H2N—CH2—CH(OEt)2
H3C—SO2Cl
⊕H Cl⊖
R R1
a OCH3 H
b O—CH2—O
c OCH3 OCH3

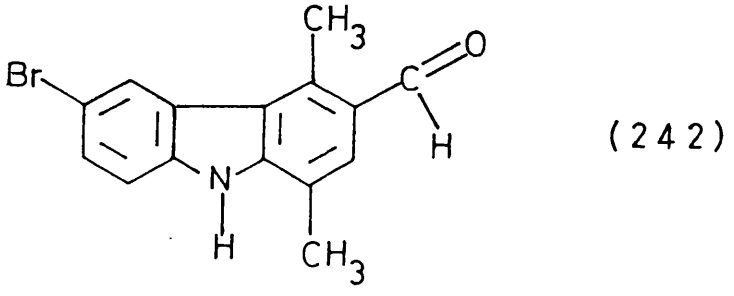
Thus 5- and 6-alkoxyl substituted indoles were condensed with hexa-2,5-dione to give the corresponding 1,4-dimethylcarbazole (234), which was formylated at the 3-position using Vilsmeier conditions to give the 3-formylcarbazole (235). Treatment of (235) with 2,2-diethoxyethylamine afforded the Schiffs base (236). This was reduced with sodium borohydride in methanol to furnish the amine (237) which on condensation with tosyl chloride furnished the corresponding N-tosyl derivative (238). Treatment of this with 6N-hydrochloric acid in dioxane at room temperature for several hours resulted in the formation of the appropriate ellipticine hydrochloride.

This route is based on the work of Jackson and Stewart[171] who reported that cyclization of the N-tosyl derivative (239) using hydrochloric acid in dioxane gave the N-tosyl-dinydroisoquinoline (240). This can be converted to the isoquinoline (241) by treatment with potassium tertiary butoxide. It is interesting to note, that application of this method to the synthesis of ellipticines results in direct aromatization to the fully aromatic tetracycles, presumably due to the greater degree of stabilization conferred by the extended conjugation in these molecules.

It could be argued that the success of this route is due to the electron releasing properties of the alkoxyl substituents however, it has been previously established by the author[19] that this route gave a 68% yield of 9-bromoellipticine when 6-bromo-3-formyl-1,4-dimethylcarbazole (242) was employed. We realised



that this result might also be partially attributed to the electron releasing ability of the bromine atom, but it did prove that this synthesis is not entirely restricted to very electron rich systems such as those used by Guthrie and his group. We were, therefore, hopeful of a satisfactory yield of ellipticine.

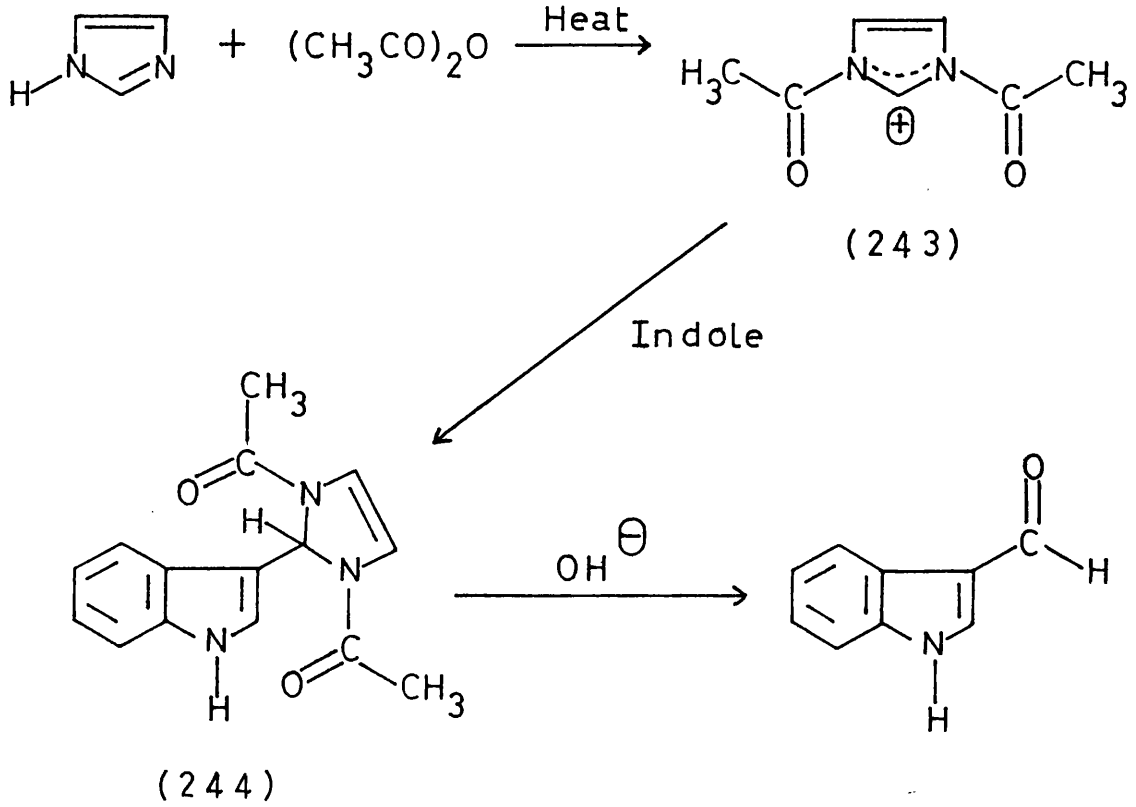


(242)

With this information to hand, we felt that only one major drawback now precluded an efficient synthesis of ellipticine, this being the problems associated with the Vilsmeier formylation of 1,4-dimethylcarbazole. This reaction gives a product that is not particularly pure, and requires steam distillation, Soxelet extraction and crystallization before it can be used for the next stage in the synthesis. The yield of pure product never exceeded 50%.

Fortunately at this juncture an interesting new formylation procedure was developed by the Swedish chemists Bergman and Sjoberg[172]. These workers found that a wide range of aromatic systems could be formylated by the use of N,N-diacylimidazolium ions. This method is outlined in (Scheme 72), for the synthesis of indole-3-carboxaldehyde from indole.

Scheme 72

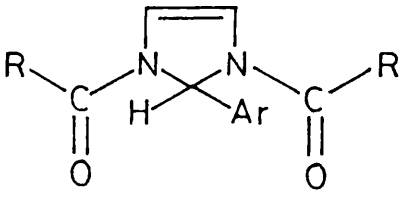


Acetylation of imidazole gives rise to the N,N'-diacetyl-imidazolium ion (243). Indole then undergoes electrophilic attack to give N,N'-diacetyl-2-(3-indolyl)-4-imidazoline (244). Hydrolysis by sodium hydroxide in aqueous ethanol then liberates indole-3-carboxaldehyde in 88% yield.

Bergman and Sjoberg[173] extended this approach to electron rich ring systems such as alkyl carbazoles, dimethoxyaryls and thiophenes, (see Table 4, for a representative range of substrates).

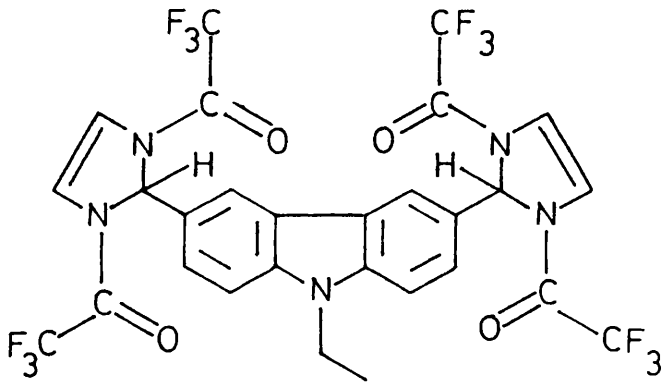
Table 4.

Ar	R	Yield %
9-Ethyl-3,6-carbazolediyl	CF_3	88
5-Methyl-2-furyl	CCl_3	48
2-Thienyl	CF_2Cl	62
4-Methoxy-1-naphthyl	CF_3	89
3,4-Dimethoxy-2-methylphenyl	CF_3	64
2-Methylindol-3-yl	CH_3	69



(244a)

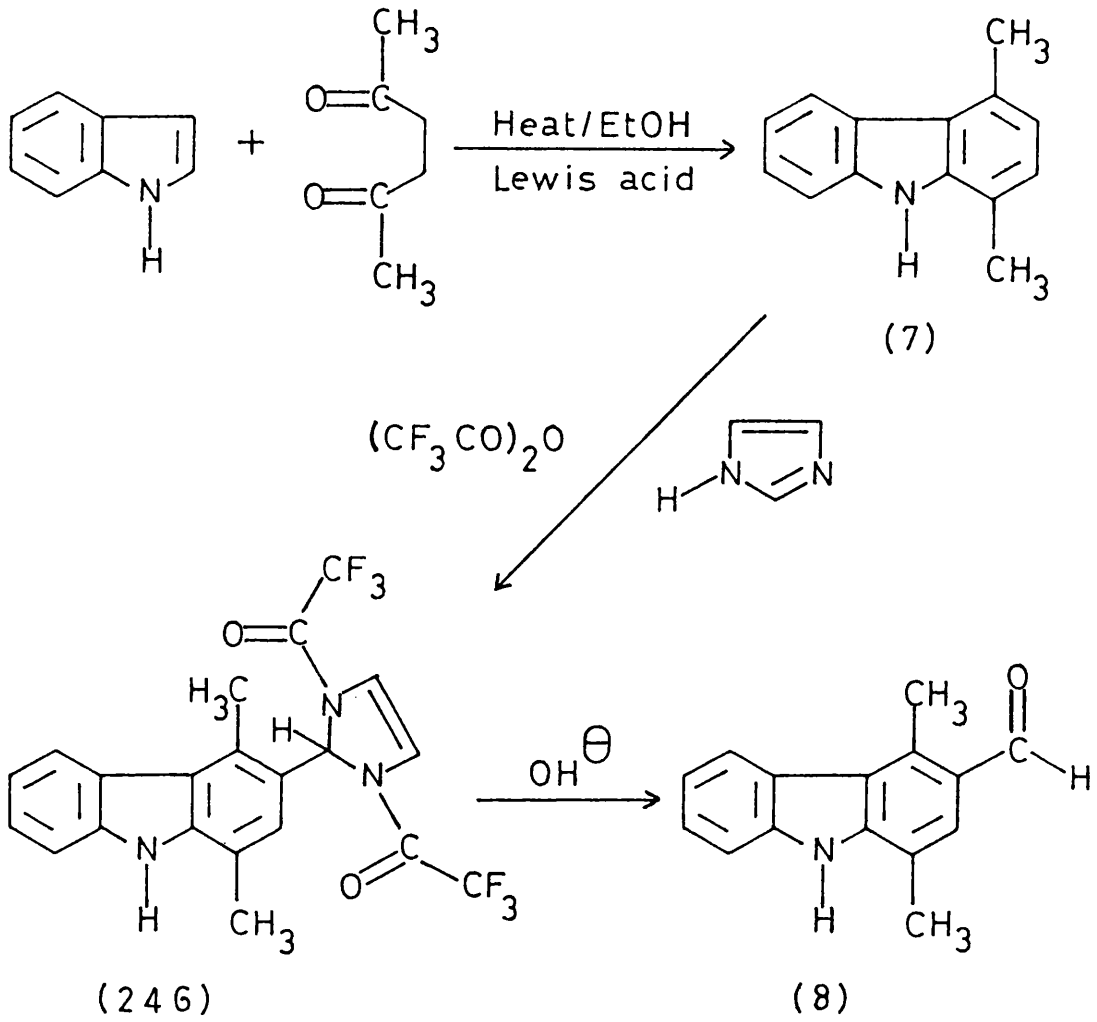
The aromatic systems that are less reactive than indole, such as methoxy naphthalenes and thiophenes require the use of the more reactive N,N-bis-(trifluoroacetyl)imidazolium ion, to give intermediates of type (244a, where R = CF_3). These reactions also require temperatures of the order of 40^{o}. We noted with interest that excellent yields were obtained with carbazoles, for example, 9-ethylcarbazole gave a 88% yield of the adduct (245).



(245)

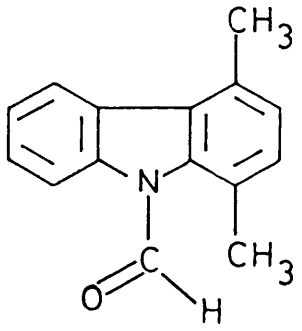
Accordingly we prepared 1,4-dimethylcarbazole by the published procedure[12] and formylated it *via* the adduct (246) as outlined in (Scheme 73).

Scheme 73.



This method gave a 77% yield of the 3-formylcarbazole (8). In line with our expectations, no 3,6-diformyl product was isolated, as the 1,4-dimethyl substitution in (7) is sufficiently electron releasing to encourage exclusive formylation at the C-3 position. This is also the case when the Vilsmeier reaction is applied to this substrate, provided an excess of the formylating agent is avoided. We did not attempt to protect the N-H position of the carbazole (7), but interestingly, none of the N-formyl product (247), was isolated. The reason for this selectivity appears to be stereochemical, as models show that the N,N-bis-(trifluoroacetyl)-

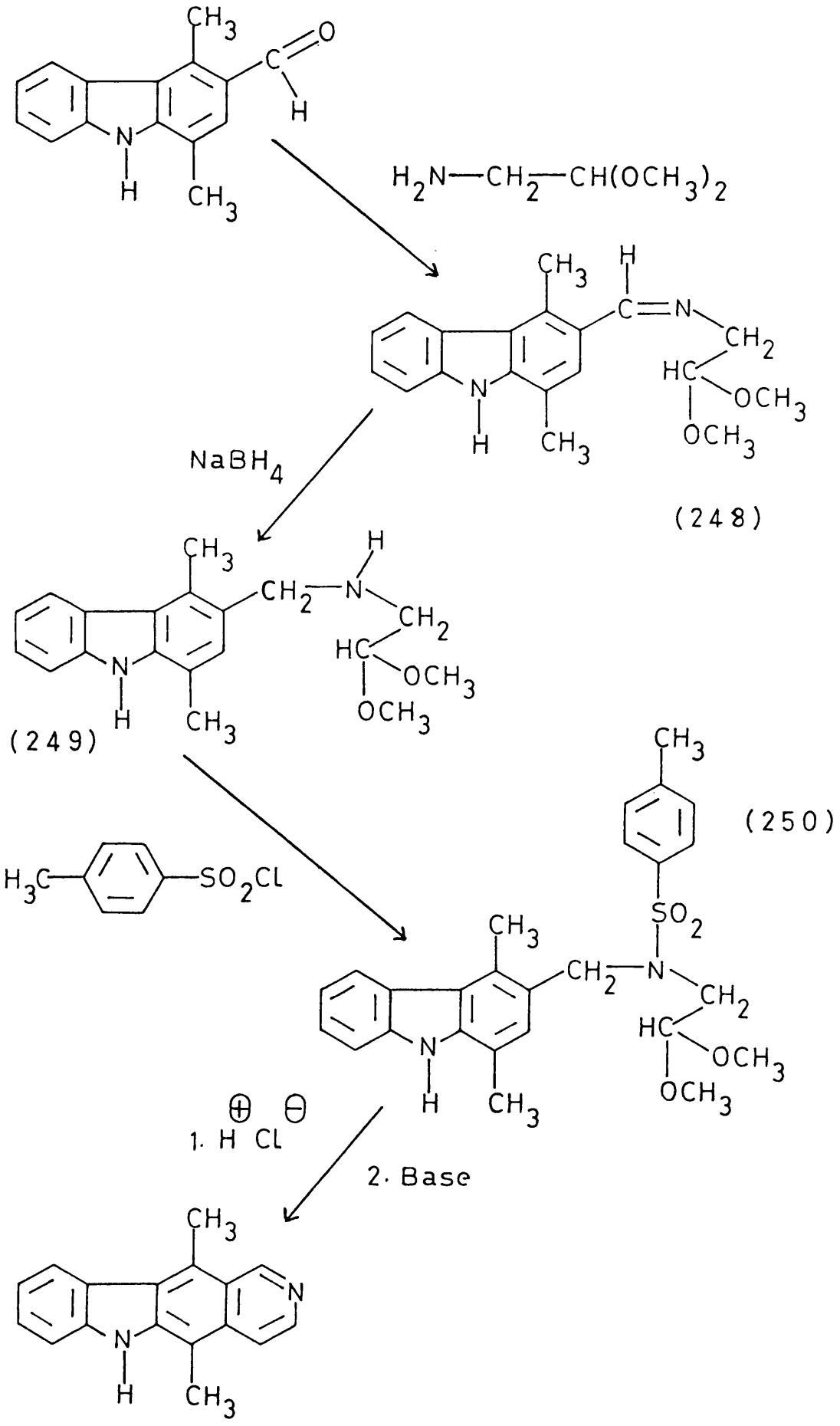
imidazolium cation would experience great difficulty in gaining access to the sterically crowded carbazole N-H position. This may be contrasted with the Vilsmeier formylation of 1,4-dimethyl-carbazole when a two molar excess of the formylating agent is employed. In this case the N-formyl compound (247) is obtained as a major by-product. Similar products have also been observed during the formylation of related carbazoles by Mathews[174] and Lallemand[56]. In this reaction the Vilsmeier intermediate (from N-methylformanilide and phosphorus oxychloride) although rather bulky is more flexible than the essentially planar N,N-bis-(trifluoroacetyl)imidazolium cation, and under forcing conditions a certain amount of N-formylation occurs.



(247)

Having obtained the 3-formylcarbazole (8), it was condensed with 2,2-dimethoxyethylamine to form the Schiffs base (248). Reduction of (248) with sodium borohydride gave the amine (249), which was N-tosylated to afford the derivative (250). Treatment of (250) with 6N-hydrochloric acid in dioxane gave a 39% yield of ellipticine after crystallization. As an additional purification step, the product was vacuum sublimed before use. This reaction sequence is outlined in (Scheme 74).

Scheme 74.

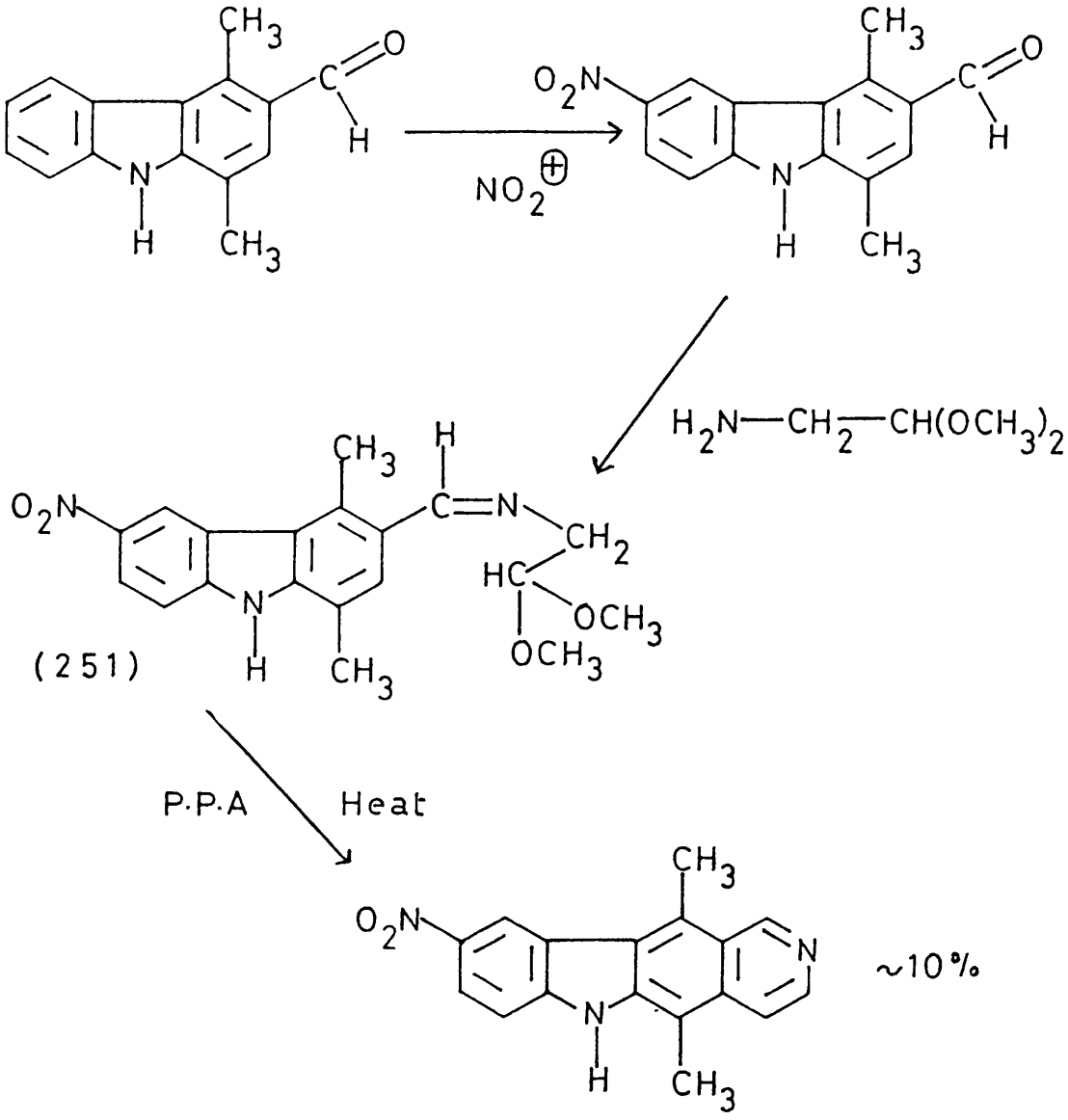


Having obtained ellipticine, we considered which substituents it would be best to employ in our preliminary investigation of the direct substitution of this alkaloid. Bromo and nitro groups were chosen as the most interesting candidates. The former because of the mildness of the conditions normally required to halogenate activated aryl rings[175], and the nitro group because we had long desired a satisfactory synthesis of nitroellipticine in order to transform it into a variety of interesting functionalities.

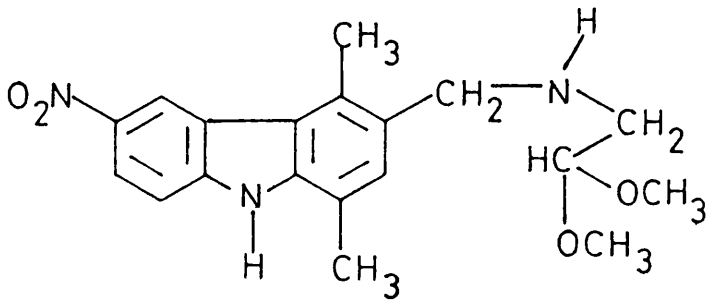
Previous attempts to prepare nitroellipticine in this laboratory had failed. Firstly, Webb[176] using a sulphuric acid/nitric acid nitrating mixture at ice-temperature obtained only intractable polymeric material, presumably due to oxidative breakdown of the ring system. A similar, but rather milder attempt with potassium nitrate in sulphuric acid at ice-temperature by Sjöberg[177] also gave tarry products that could not be purified. A third approach to this problem was made at an earlier time by the author[19]. This work established that in accordance with Dalton's[12] findings attempts to ring close the nitro Schiffs base (251) with hot 99-100% ortho-phosphoric acid gave very low yields($\sim$ 10%) of 9-nitroellipticine in an insufficiently pure state for full characterisation. This work is outlined in (Scheme 75).

In a bid to improve this ring closure, reduction of the C=N bond of (251) was attempted, using sodium borohydride as before. It was hoped that this would give the expected product

Scheme 75.

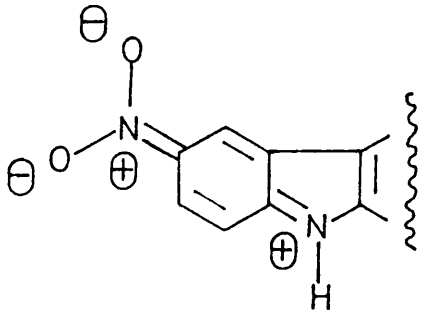


(252), and hence access to the milder conditions of (Scheme 74, p. 195). However, only a low yield of (252) was obtained, accompanied by a multicomponent mixture, resulting from the partial reduction of the nitro group.



(252)

The result of this experiment seemed rather unusual at first sight, as it is normally claimed[178] that nitro groups are inert to reduction by sodium borohydride. However, it is thought that in this case the nitro group is made more susceptible to reduction by the presence of the secondary NH group in ring 'B'. As this is situated <u>para</u> to the nitro group it can exercise a strong mesomeric influence, possibly by contributions from the form (253).

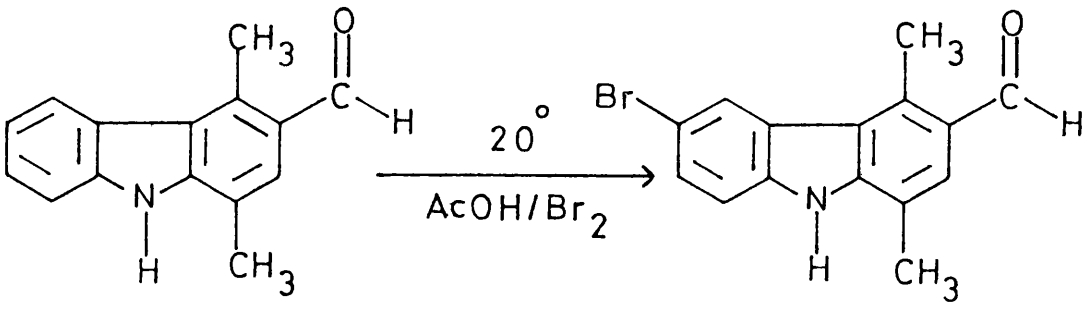


(253)

The form (253) must be of fairly high energy and it is thought that the drive to re-form the normal aromatic structure must contribute to the ease of reduction. Partial reduction of similar 6-nitrocarbazoles by sodium borohydride has been observed in this laboratory by Webb[179], during the course of a related study.

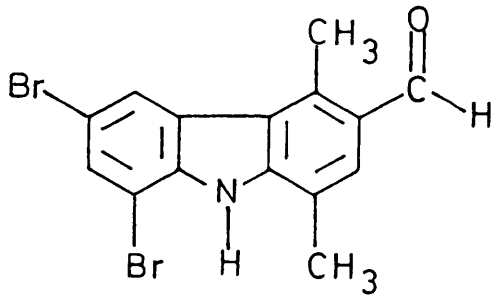
In view of these difficulties, work on designing a synthesis with the nitro group built in was abandoned. We also felt that methods involving nitrated carbazole precursors would not give rise to a favoured ring closure reaction due to the deactivating effect of the nitro group on the aromatic system.

We decided to attempt the bromination of ellipticine first, and the method of choice was similar to that used by Dalton[12] to successfully brominate the 3-formyl carbazole (8) in the C-6 position.



(8) (242)

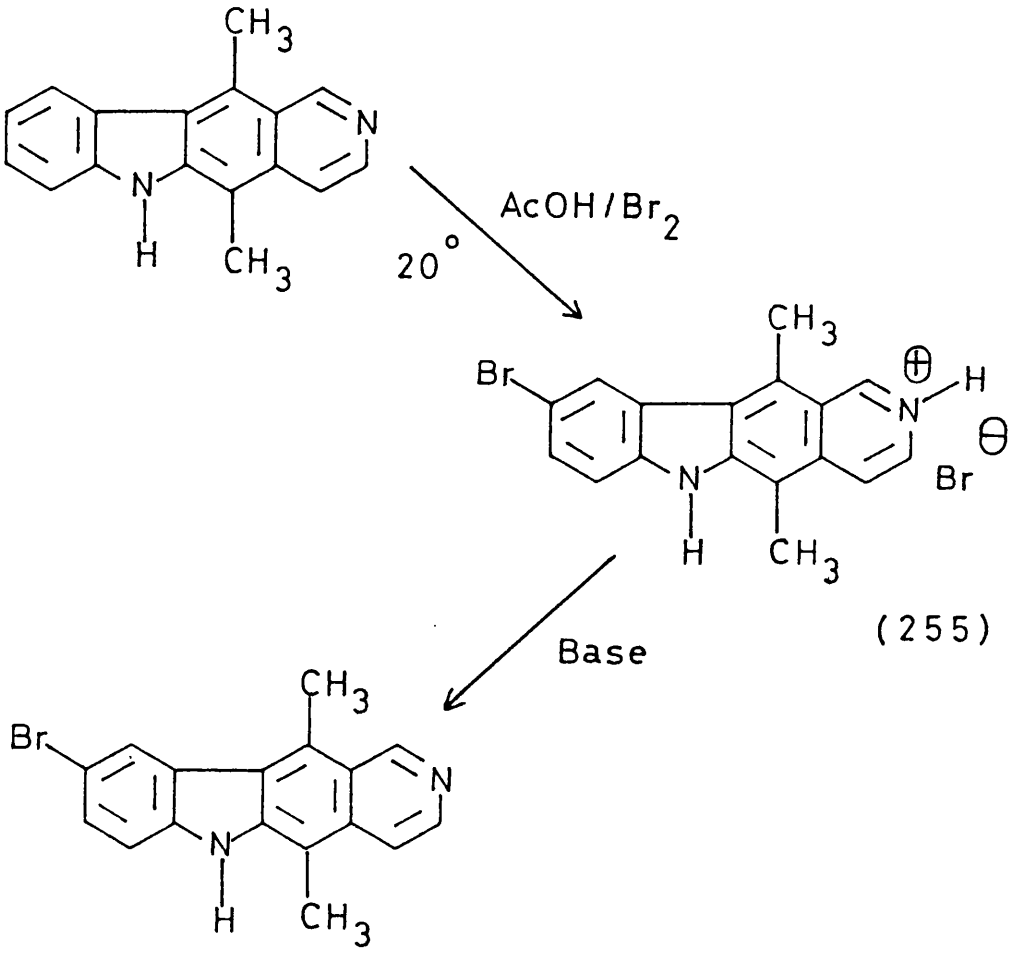
The reaction is performed in glacial acetic acid containing one mole equivalent of bromine at room temperature, under these conditions an immediate precipitate of (242) is obtained, in 92% yield after crystallization. Previous experience[19] has shown that an excess of bromine must be avoided, or the 6,8-dibromo compound (254) contaminates the product.



(254)

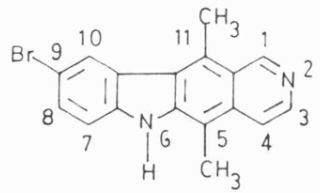
Under the very mild conditions of this experiment it is to be expected that the bromine atom will enter the most electronegative carbon site, <u>para</u> to the ring 'B' nitrogen atom in the unsubstituted 'A' ring. It is well established that the C-6 and C-8 positions are those most likely to brominate in a carbazole system 'A' ring. 3-Bromo, 3,6-dibromo, 1,3,6-tribromo and 1,3,6,8-tetrabromo carbazoles being obtained depending on the quantity of bromine used[180]. 3,6-Dibromo and 1,3,6-tribromocarbazoles were obtained by Mizuch and Sanchenko[181], similar results have been observed by Lindermann and Muhlhaus[182] and a number of other groups[183,184]. Coulson and Longuet-Higgins[185] have carried out electron density calculations which support these experimental results.

On carrying out the reaction with ellipticine an immediate bright yellow precipitate was obtained, presumably the salt (255), which was dissolved in water and basified with sodium hydrogen carbonate, whereupon a second yellow precipitate was collected and crystallized from ethanol to give a 93% yield. This product proved to be identical in every way with a sample of 9-bromo-ellipticine prepared independently by the method Guthrie and his co-workers[170] have employed, (see p. 186).



The ultraviolet and infrared spectra clearly show that bromination has occurred and mass spectral analysis proves it to be monobromination. Full supporting spectral data are included in the experimental section (see p. 345).

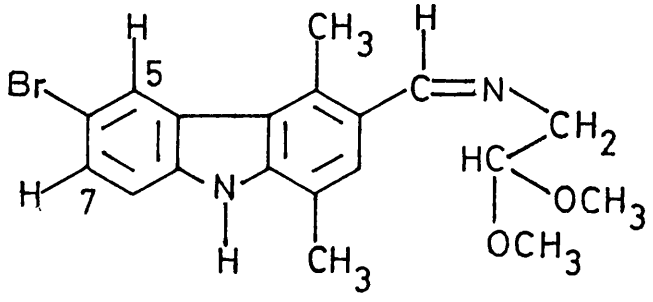
(Figure 6)



1H occurs as a s, at 9.87

The 200 MHz ^{1}H n.m.r. spectrum in d_6 D M.S.O. displays signals for the 'A' ring as follows: 7.45 (1H, d J 8Hz-7H), 7.63 (1H, d x d J 8Hz and 2Hz-8H) and 8.27 (1H, d J 2Hz-10H) p.p.m. The rest of the spectrum is consistent with that expected for 9-bromoellipticine and it is reproduced in (Fig. 6). The ^{1}H n.m.r. data indicate that the bromine atom has entered the C-9 position. The A B X pattern rules out the possibility that the product is 7-bromoellipticine, and the downfield shift of the signals assigned to the C-8 and C-10 protons, with respect to ellipticine itself (where they occur above 7.4 p.p.m.), indicate. that these two positions are adjacent to the bromine atom, which is exerting its electron withdrawing effect.

It is also instructive to compare the positions of the C-8 and C-10 proton signals with those obtained for the equivalent C-7 and C-5 protons of the compound (256), measured at a previous time in this laboratory.[19]

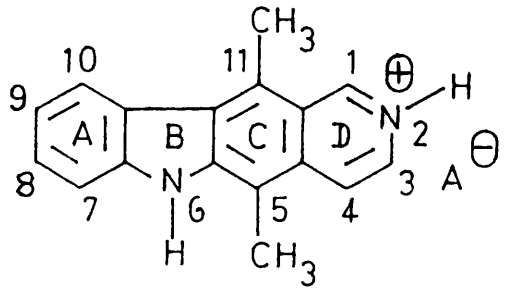


(256)

Here, the values are: ($CDCl_3$) 7.50 (1H, d x d J 8Hz and 2Hz-7H) and 8.10 (1H, d J 2Hz - 5H) p.p.m. This can be compared with $\delta[(CD_3)_2SO]$ 7.58 (1H, d x d J 8Hz and 2Hz - 8H) and 8.22 (1H, d J 2Hz - 10H) for the product.

Allowing for the difference in the structure of this model compound and the fact that the values were measured in different solvents, it can be seen that they show a close correlation. Further more, electron density calculations for ellipticine[186], indicate that the two positions most favoured for electrophilic attack in the 'A' ring are at C-9 and C-7. The likely order of reactivity is also predicted to be C-9 C-7, in accordance with carbazoles in general. The other physical data, including the melting point of 317-319°, agree with the results obtained for this compound by Dalton and his group[12].

Despite the data quoted above the possibility exists that the salt (257) may form prior to bromination of the 'A' ring.

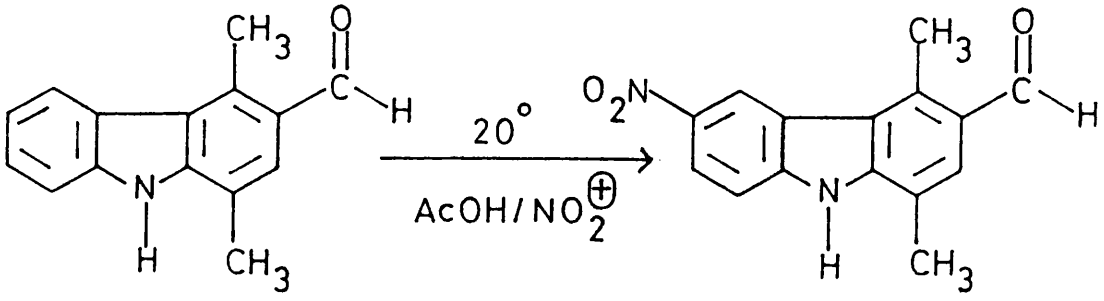


(257)

If this were the case, the positively charged ring 'D' nitrogen atom would tend to decrease the electron availability at the ring 'B' nitrogen and cause a deactivating effect in the 'A' ring. The situation in this ring would then more resemble that in the anilinium cation, and it is conceivable that bromination might occur in the C-8 position. As both derivatives give rise to an A B X pattern it is difficult to specify with certainty which has been formed on the basis of the ^{1}H n.m.r. evidence alone.

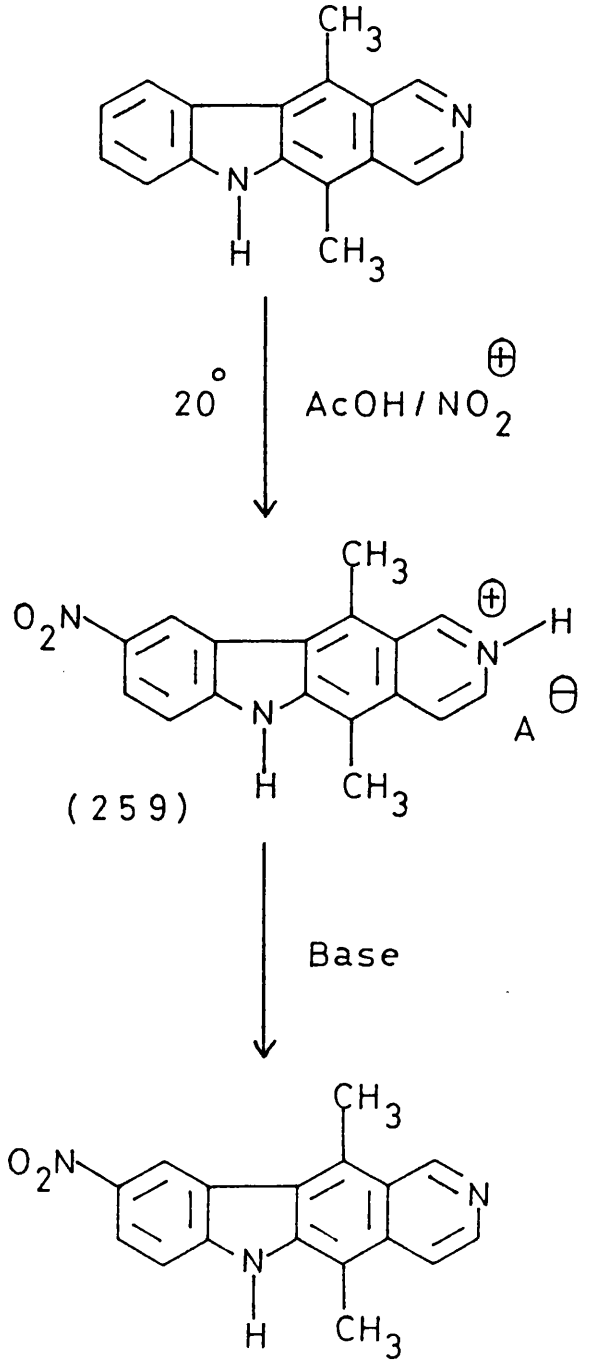
Due to this slight possibility for error in our interpretations we have submitted a sample of the product for X-ray diffraction analysis, and the results are awaited with interest.

Encouraged by the success of this approach with bromoellipticine we sought to apply a similar technique to the preparation of nitroellipticine. Dalton and his group[12], have also successfully nitrated the 3-formylcarbazole (8) using a mixture of acetic acid, nitric acid and sulphuric acid in a ratio of 93:6:1 at room temperature to give the 6-nitro derivative (258) in good yield.



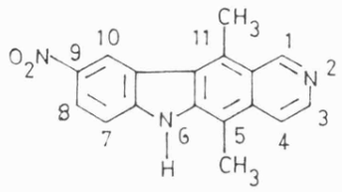
(8) (258)

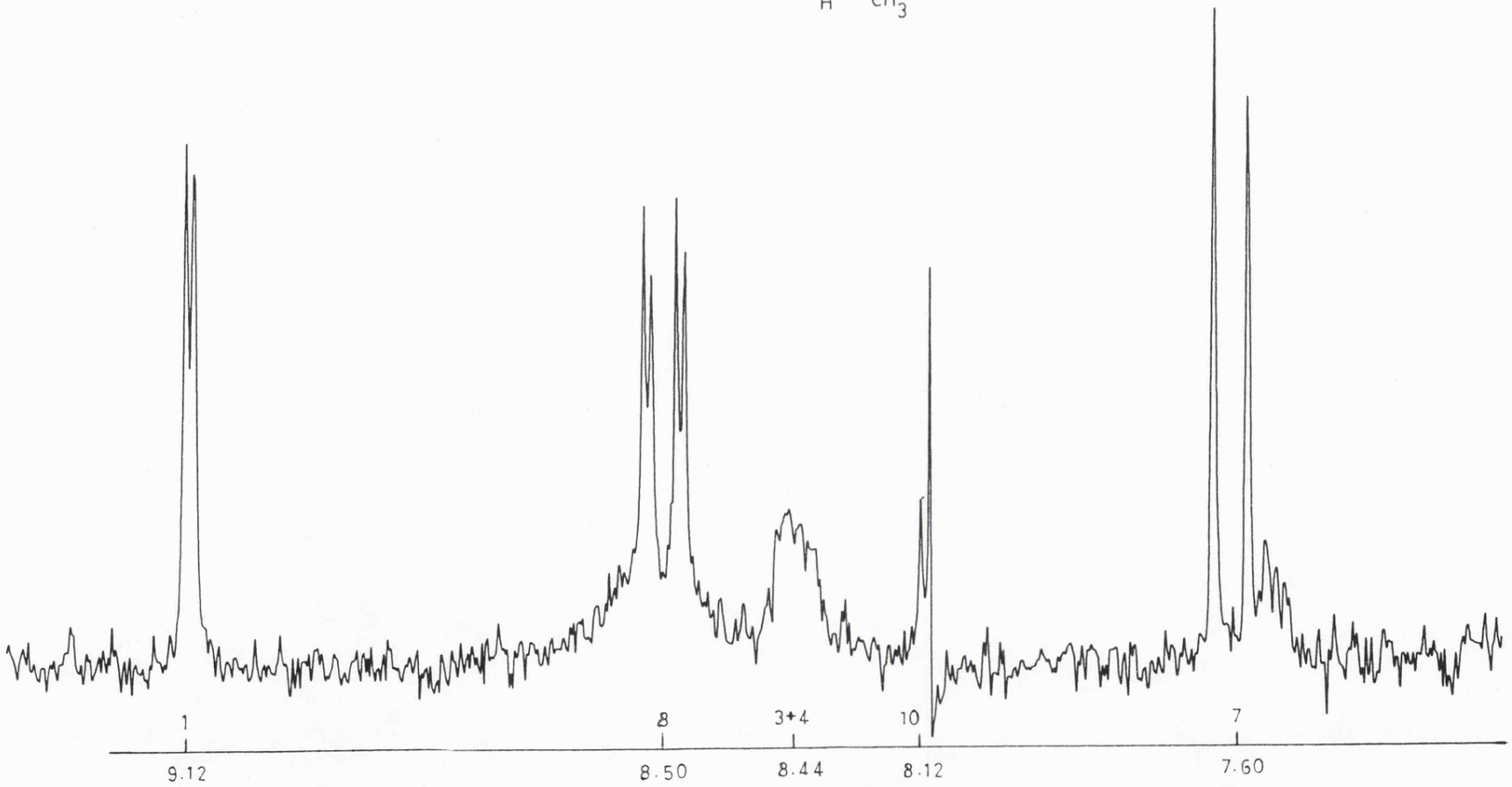
We used the same mild method for ellipticine, and after a short induction period, obtained a bright yellow precipitate, presumably the salt (259). This was dissolved in water and basified with sodium hydrogen carbonate to give an amorphous yellow solid, which was crystallized from glacial acetic acid as yellow prisms in 52% yield.



(Figure 7)

Aromatic region X 10

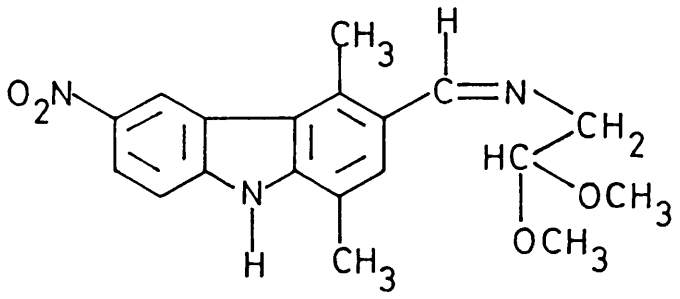




The ultraviolet spectrum showed the usual absorptions associated with the ellipticine skeleton and a band at $\lambda(\varepsilon)$ 387(14,600)nm due to the extended conjugation of the nitro group. The infrared spectrum supports this with absorptions due to the nitro group at(1545 antisymmetric stretching) and (840 symmetric stretching)cm^{-1}. The 200 MHz ^{1}H n.m.r. spectrum in d_6 D.M.S.O. displays signals for the 'A' ring as follows: δ 7.60 (1H, d J 8Hz-7H), 8.12 (1H, d J 2Hz-10H), 8.50 (1H, d x 2 J 8Hz and 2Hz-8H), and is reproduced in (Figure 7). The rest of the spectrum is entirely consistent with what is to be expected for a 9-substituted ellipticine structure. The mass spectrum shows a molecular ion, base peak at a mass to charge ratio of m/e$^{+\cdot}$ 291(100) and another significant peak occurs at m/e 245, corresponding to the simple fission of NO_2, supported by a metastable ion at m/e 206.3.

Once again it is obvious from the spectral data that the 'A ring of ellipticine has undergone mono-substitution. A study of the ^{1}H n.m.r. spectrum at 200 MHz defines the position of nitration and rules out 7-nitroellipticine. As expected all the protons of the 'A' ring display a downfield shift and the signals due to the C-10 and C-8 protons show that these positions are deshielded to a significant extent with respect to those of ellipticine itself. The N-H proton resonates at 12.5 p.p.m. compared with 11.5 p.p.m. for ellipticine itself, and this 1.0 p.p.m. downfield shift supports the conclusion that the nitro group is sited in a para relationship to the N-H, where it can exert its full mesomeric, electron withdrawing influence.

Again it is useful to compare the values obtained for the C-10 and C-8 protons of the product with those for the equivalent C-5 and C-7 protons of the compound (251) measured at a previous time in our laboratory.



(251)

Here, the values are: δ ($CDCl_3$) 7.90 (1H, d J 2Hz-5H) and 8.25 (1H, d x d J 8Hz and J 2Hz-7H) p.p.m. This can be compared with $\delta[(CD_3)_2SO]$ 8.12 (1H, d J 2Hz-10H) and 8.50 (1H, d x d J 8Hz and J 2Hz-8H) p.p.m. for the product.

As the Schiffs base (251) is derived from the appropriate carbazole by nitration, the position of substitution is not in doubt, as carbazole nitrations are well documeted. Thus, Lindemann[187], Ruff and Stein[188] and Tucker and Stevens[189] have shown that the 3- and 6-positions are most reactive towards nitrating agents and the 1- and 8-positions somewhat less so. Exhaustive nitration gives 1,3,6,8-tetranitrocarbazole[190].

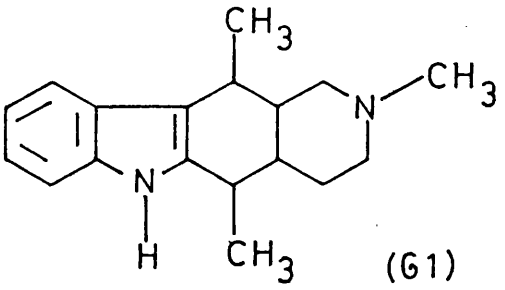
Thus, allowing for the differences in structure and solvent the values are in sufficiently close agreement to indicate that the nitro group occupies the C-9 position of the ellipticine structure.

Other physical data, including the very high melting point of 358° agree with the results obtained by Dalton[12] and Gansser et al[191], who both used modifications of Cranwell and Saxton's[8] original route (see p. 6).

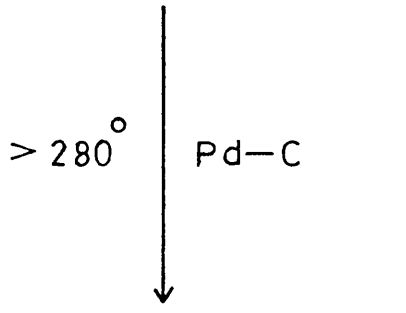
Thus, we have shown that it is possible to prepare 9-bromo and 9-nitroellipticines by direct electrophilic substitution of the 'A' ring. The advantages of this approach are that it is very much easier to synthesize ellipticine itself than many of its derivatives and the process is rapid, clean and reasonably efficient. This technique is to be recommended for the synthesis of simple derivatives rather than the implementation of more elaborate and less direct methods for which rather inaccessible starting materials are required. 9-Nitroellipticine in particular, has potential for conversion into a wide range of other C-9 derivatives via its reduction product 9-aminoellipticine.

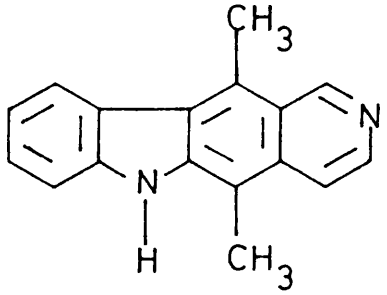
Having successfully pioneered the first synthesis of 8-hydroxyellipticine we considered ways of improving the synthesis so that (1.0g - 500mg) quantities might be prepared for pharmacological and clinical evaluation. Due to the problems associated with the synthesis of 6-methoxyindole and the subsequent Grignard condensation with haloethylpyridines discussed earlier, we sought a method that did not involve the use of indole precursors.

It has long been recognized that one of the main problems associated with Stillwells synthesis of ellipticine[11] (see p. 8), is the severity of the final, high temperature dehydrogenation step required to aromatize the N-methyl octahydroellipticine (61).



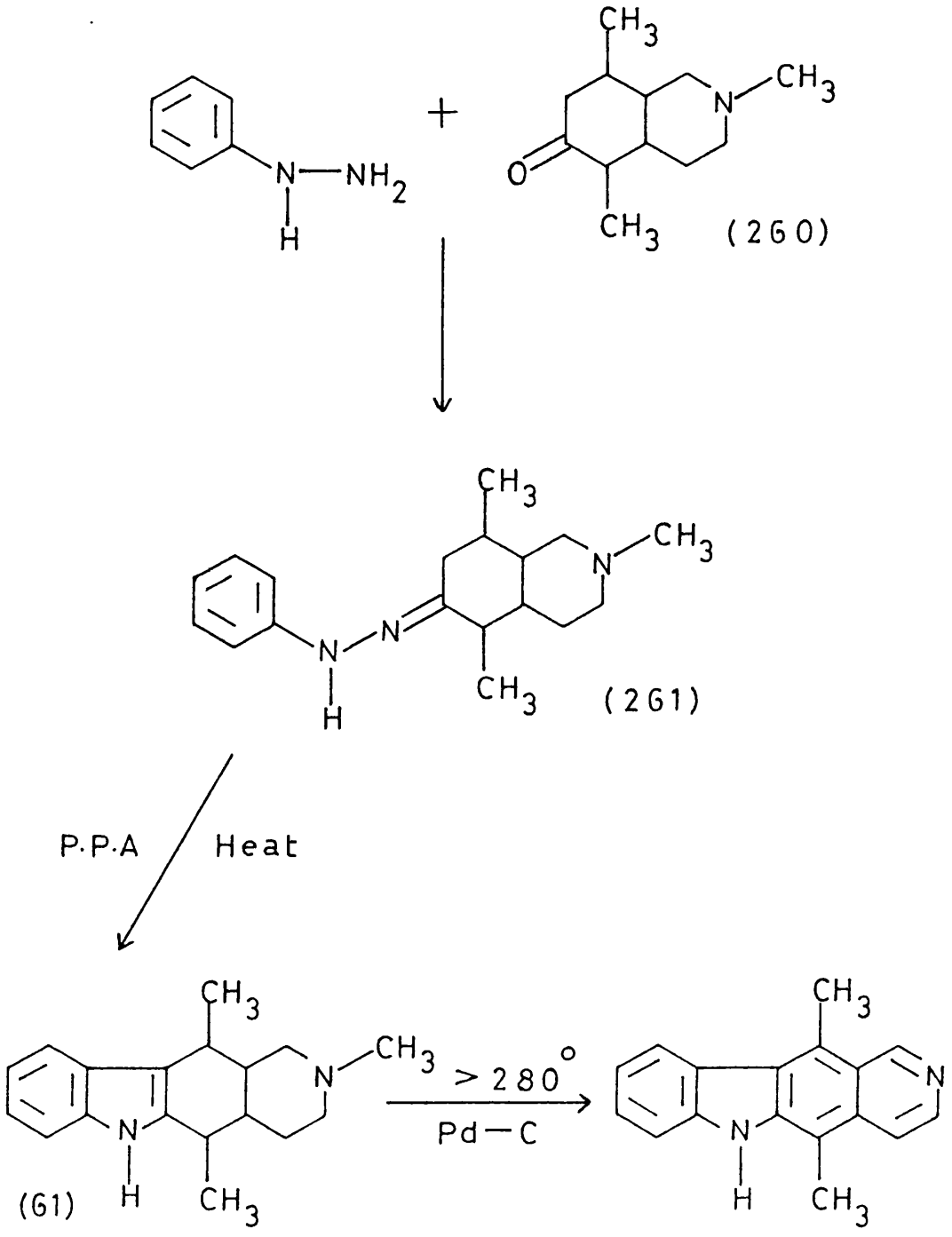
(61)





This compound is prepared by a Fischer indole condensation of octahydro-2,5,8-trimethyl-6(2H)-isoquinoline (260) with phenylhydrazine to give the hydrazone (261), which is then ring closed in hot polyphosphoric acid (P.P.A.) to afford (61), as outlined in (Scheme 76).

Scheme 76.

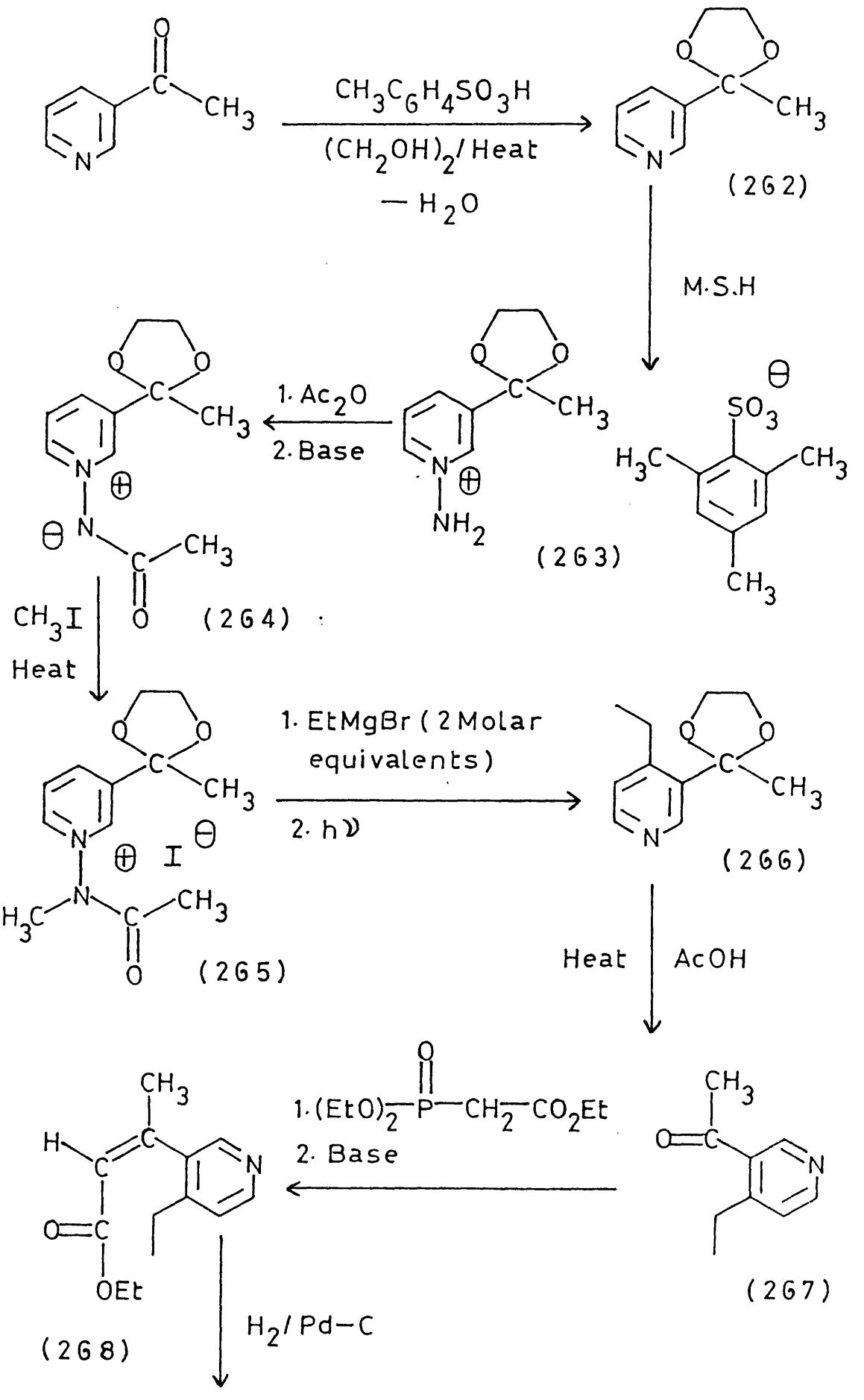


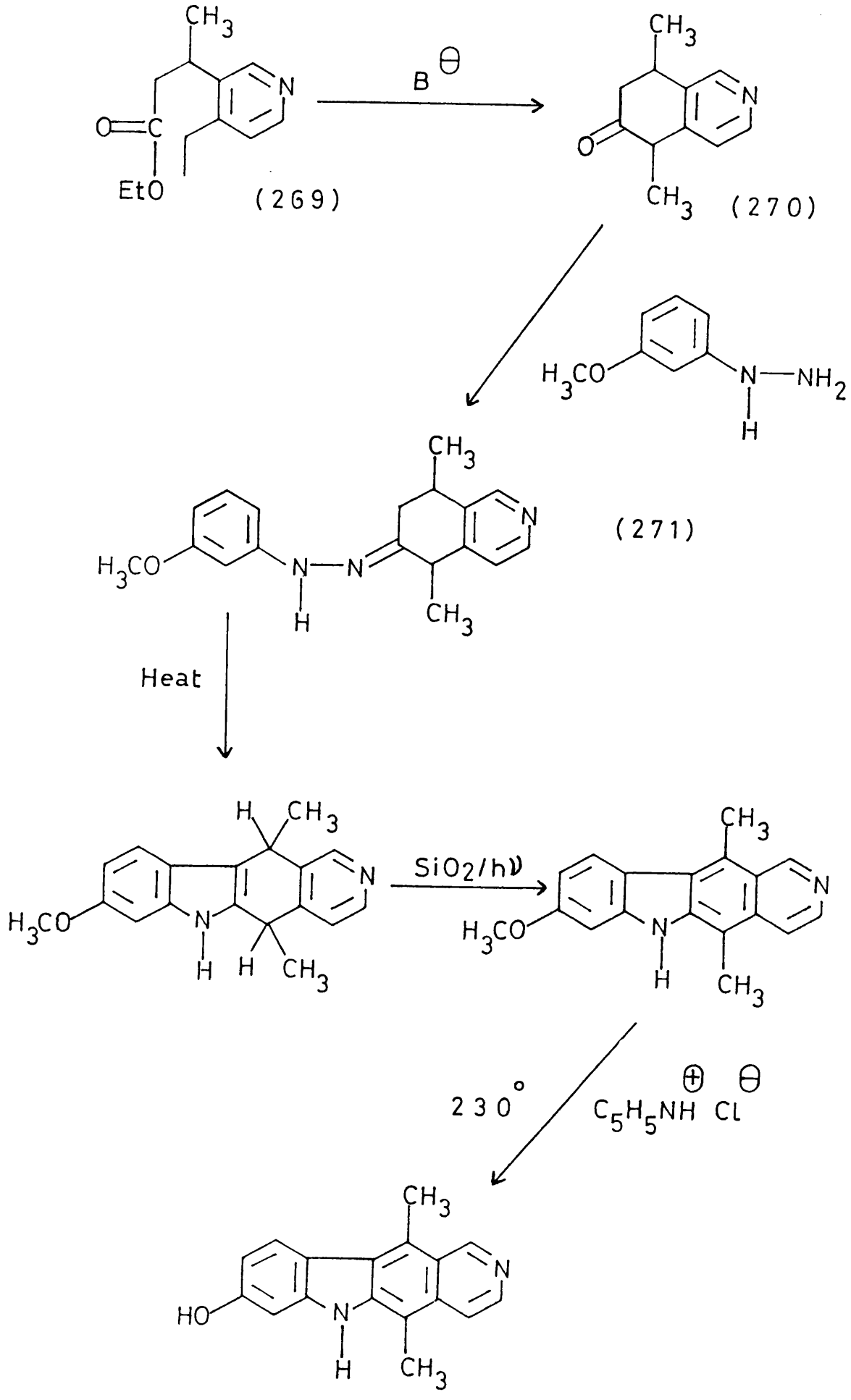
We felt that if the pyridine analogue (270) , (Scheme 77) of the piperidine compound (260), could be prepared it would be a much more satisfactory substrate for the remainder of the synthesis. Hence condensation with phenylhydrazine, followed by Fischer indolization would then give rise to 5,11-dihydro-ellipticine, which can be aromatized under mild conditions as described previously, (see p. 19). Furthermore, various arylsubstituted hydrazines could be introduced to make the method entirely flexible. Thus, 3-methoxyphenylhydrazine would give 8-methoxy and hence 8-hydroxyellipticine.

3-Acetylpyridine can be easily acetalysed to provide the cyclic acetal (262). This can be converted to the methiodide salt (265) in a similar manner to that previously described for compounds (35) and (228), (see pp. 23 and 181). Thus mesitylene sulphonylhydroxylamine (M.S.H.) gives the pyridinium salt (263). Treatment of this with acetic anhydride, followed by the addition of base affords the Zwitter ionic acetylimide (264), which on methylation with methyl iodide furnishes the bright yellow methiodide (265).

Our synthetic plan then required reaction of the iodide (265) with ethylmagnesium bromide[192] under anhydrous conditions followed by irradiation of the product in ethanol with ultraviolet light. We anticipate that this would furnish the 4-ethylcompound (266). Treatment with acid would then provide the 4-ethylpyridine

Scheme 77.


CH3C6H4SO3H
(CH2OH)2/Heat
— H2O
(262)
M.S.H
1. Ac2O
2. Base
(263)
(264)
CH3I
Heat
1. EtMgBr (2 Molar equivalents)
2. hν
(265)
(266)
Heat
AcOH
1. (EtO)2P—CH2—CO2Et
2. Base
(267)
(268)
H2/Pd—C


CH3
N
O
C
EtO
B ⊖
(269)
CH3
N
O
CH3
(270)
H3CO
N—NH2
H
CH3
N
H3CO
N—N
H
CH3
(271)
Heat
H
CH3
N
H3CO
N
H
H
CH3
SiO2/hν
CH3
N
H3CO
N
H
CH3
230°
⊕ ⊖
C5H5NH Cl
CH3
N
HO
N
H
CH3

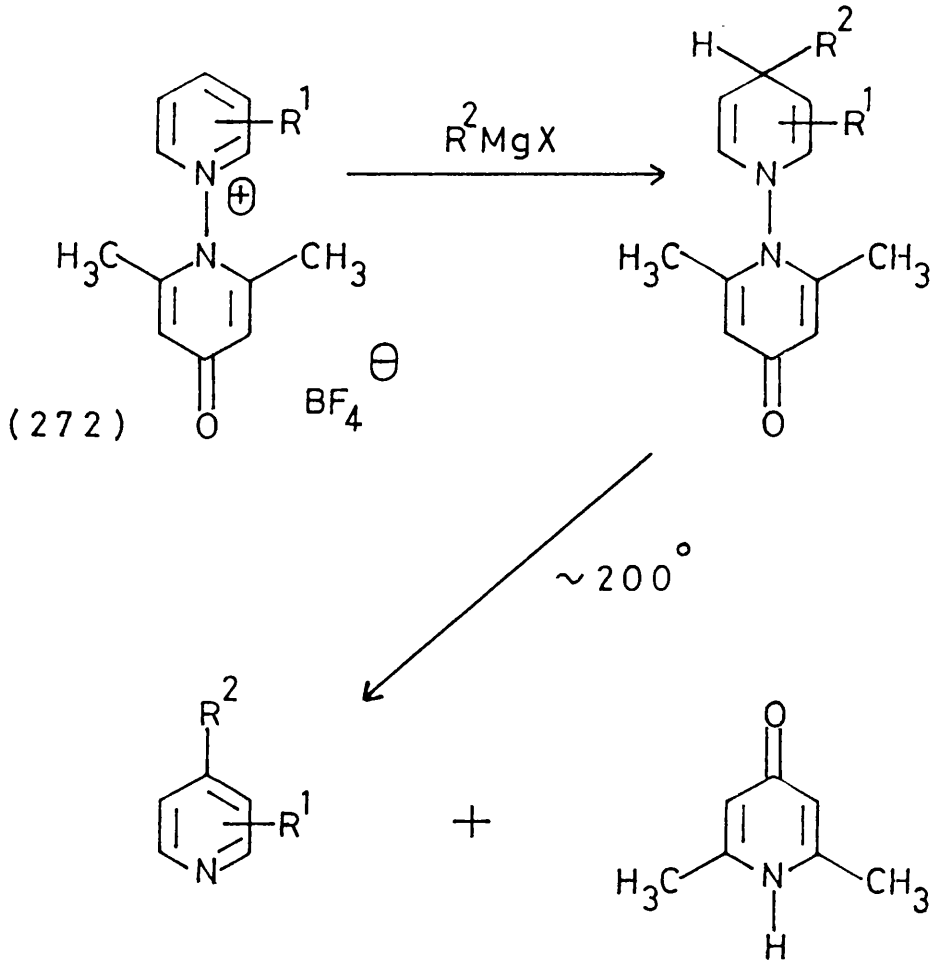
(267). A Horner-Emmon's reaction on the carbonyl group of (267) using triethylphosphonoacetate in base would give the α,β-unsaturated ester (268) and hydrogenation over palladium on carbon would then afford the ester (269), which on treatment with base may ring close to the cyclic pyridylketone (270).

Condensation of (270) with 3-methoxyphenylhydrazine would then give the hydrazone (271), which if subjected to the conditions of the Fischer indolisation reaction would afford 5,11-dihydro-8-methoxyellipticine. This could easily be aromatized by exposure to ultraviolet light on a silica support to furnish 8-methoxyellipticine. Finally, this could be O-demethylated[62], to give 8-hydroxyellipticine.

The key step in this sequence is the insertion of an ethyl group into the 4-position of the pyridinium salt (265), using a Grignard reagent. In line with our previous experience (see p 181), we hoped that the bulky N-methylacetamido group on the pyridine nitrogen atom of (265) would serve a dual function. Firstly to sterically shield the α-positions of the ring from attack, and secondly to promote reaction of the nucleophile at the γ-position. Furthermore, its properties as a good leaving group assists the reaction by virtue of its ready expulsion from the first formed 1,4-dihydropyridine intermediate.

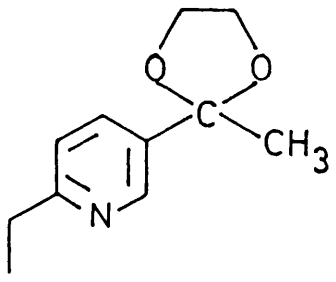
At this time we read with interest a recent paper by Katritzky and Sammes[192], who employed a similar methodology to prepare a variety of 4-substituted pyridines by attack of Grignard

reagents on N-(2,6-dimethyl-4-oxopyridin-1-yl)pyridinium salts such as (272). This approach has since been extended to other nucleophiles[193-195].



Accordingly, we prepared the acetal (262) in the published manner[18], and treated it with the series of reagents previously described (p. 211), to give the salt (265) in 96% yield.

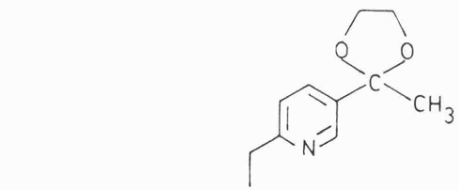
Unfortunately the reaction of this methiodide salt (265) with two molar equivalents[192] of ethylmagnesium bromide under anhydrous conditions, did not give rise to the 4-ethylpyridine (266), as hoped, but rather, the 2-ethylpyridine (273).

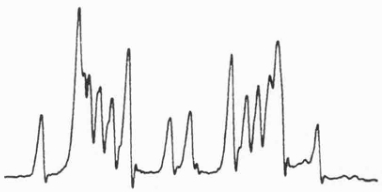


(273)

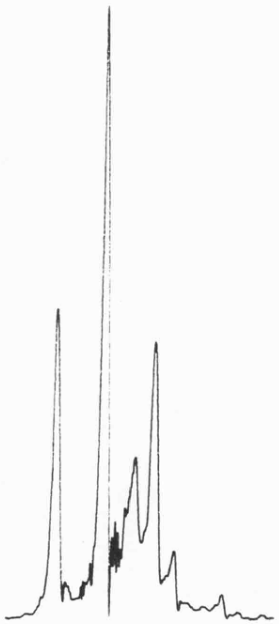
The aromatic region of the ^{1}H n.m.r. of the product (273) is reproduced in (Figure 8), and for comparison purposes the spectrum of the 3,4-disubstituted pyridine (41) accompanies it. The protons at the α positions of the pyridine ring always occur at lower field than those at other sites due to the electron withdrawing effect of the adjacent nitrogen atom, but in 3,4-disubstituted derivatives the resonances due to the C-2 and C-6 protons form a characteristic pattern. The 2H proton appears as a doublet with a J value of 2Hz and the 6H proton as a double doublet with J values of 8 and 2Hz respectively. In general, these signals occur close together at low field, as can be seen from (Figure 9) for the 3,4-disubstituted pyridine (41), for which the chemical shift positions are: δ ($CDCl_3$) 7.35 (1H, d J 7Hz-5H), 8.60(1H, d x d J 8Hz and 2Hz - 6H) and 8.90 (1H, d J 2Hz-2H).

(Figure 8)

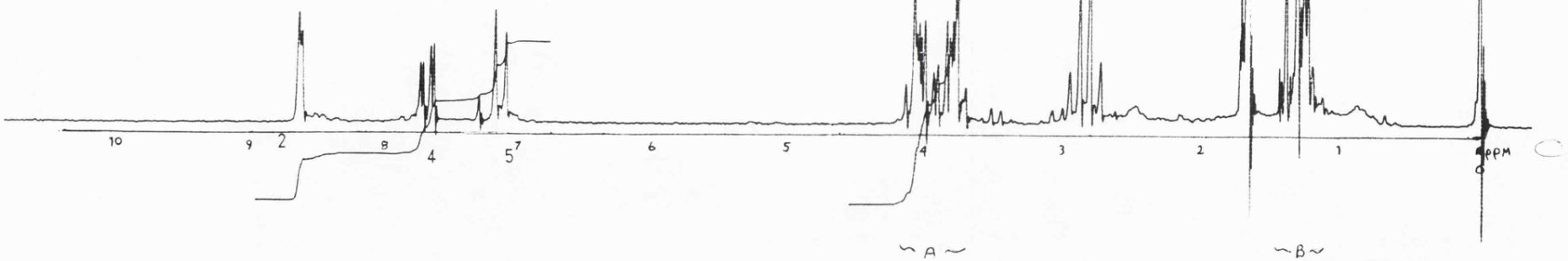




A × 4

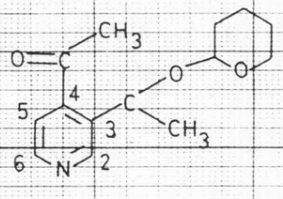


B × 4

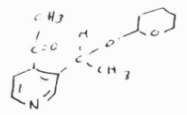


~A~ ~B~

(Figure 9)



SPECTRUM NO. DM 10
DATE 17-6-75
FREQ.
NUCLEUS 1H
SAMPLE



SOLVENT	$CDCl_3$
CONC.	5 mgs
REFERENCE	TMS
LOCK	EXT
TEMP.	28 °C
R. F. LEVEL	40
A.F. LEVEL ANALYTICAL	3-5
LOCK	
SD	
AMPLITUDE ANALYTICAL	X10-10
LOCK	
INTEGRATOR	
FILTER	20
OFFSET	H PPM

FREQ. FIELD/FREQ. (FIELD.)

OPERATOR D. Wood

REMARKS:

No change on deuteration.

SWEEP TIME SEC.

25	50	100	(250)	500
1000	2500	5000	10000	

SWEEP WIDTH Hz X0.01PPM

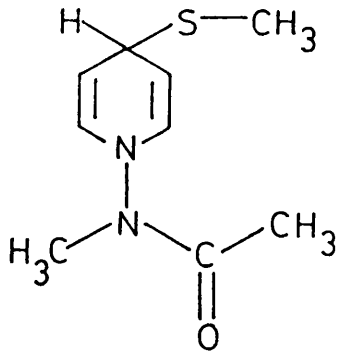
27	54	108	270	540
(1080)	2700	5400	10800	

WIDE SWEEP GAUSS

10.8	27	54	108	540

This characteristic pattern is absent from the spectrum of the product (273), for which the data are: δ ($CDCl_3$) 7.45(1H, d J 7Hz-5H), 8.15 (1H, d x d J 7Hz and 2Hz-4H) and 9.10 (1H, d J 2Hz-2H), indicating that the ethyl group has entered the C-6 position rather than the required C-4 site.

The reason for this unexpected substitution may be that the cyclic acetal group at the C-3 position is too bulky and rigid to allow the attack of the Grignard reagent at the adjacent site, and hence, the reaction is directed to the relatively unhindered C-6 position. This view is supported by an examination of models and it may be significant that the largest group employed in the C-3 position by Katritzky and Sammes[192] in their successful C-4 substitutions was a methyl function. Lyle and Gauthier[196] have established that 1,4-dihydropyridine intermediates are more thermodynamically stable than the 1,2-isomers, though the latter are usually formed from pyridinium salts under kinetic control. Katritzky and Sammes[195] speculate that any 1,2-dihydrointermediate formed from a substrate bearing an N-N linked protecting group that does not fully shield the 2- positions of the pyridine ring, might subsequently rearrange to the 1,4-dihydro isomer, and a similar effect has been observed with cyanide ion as the nucleophile[197], although this is in conflict with the results of related reactions carried out in this laboratory. Okamoto and his co-workers[198] have isolated only the 1,4-dihydro-intermediate (274) from the reaction between methanethiolate anion and 1-(N-methylacetamido)pyridinium iodide, again this is thought to be due to rearrangement of any 1,2-dihydro-intermediates formed.[195]



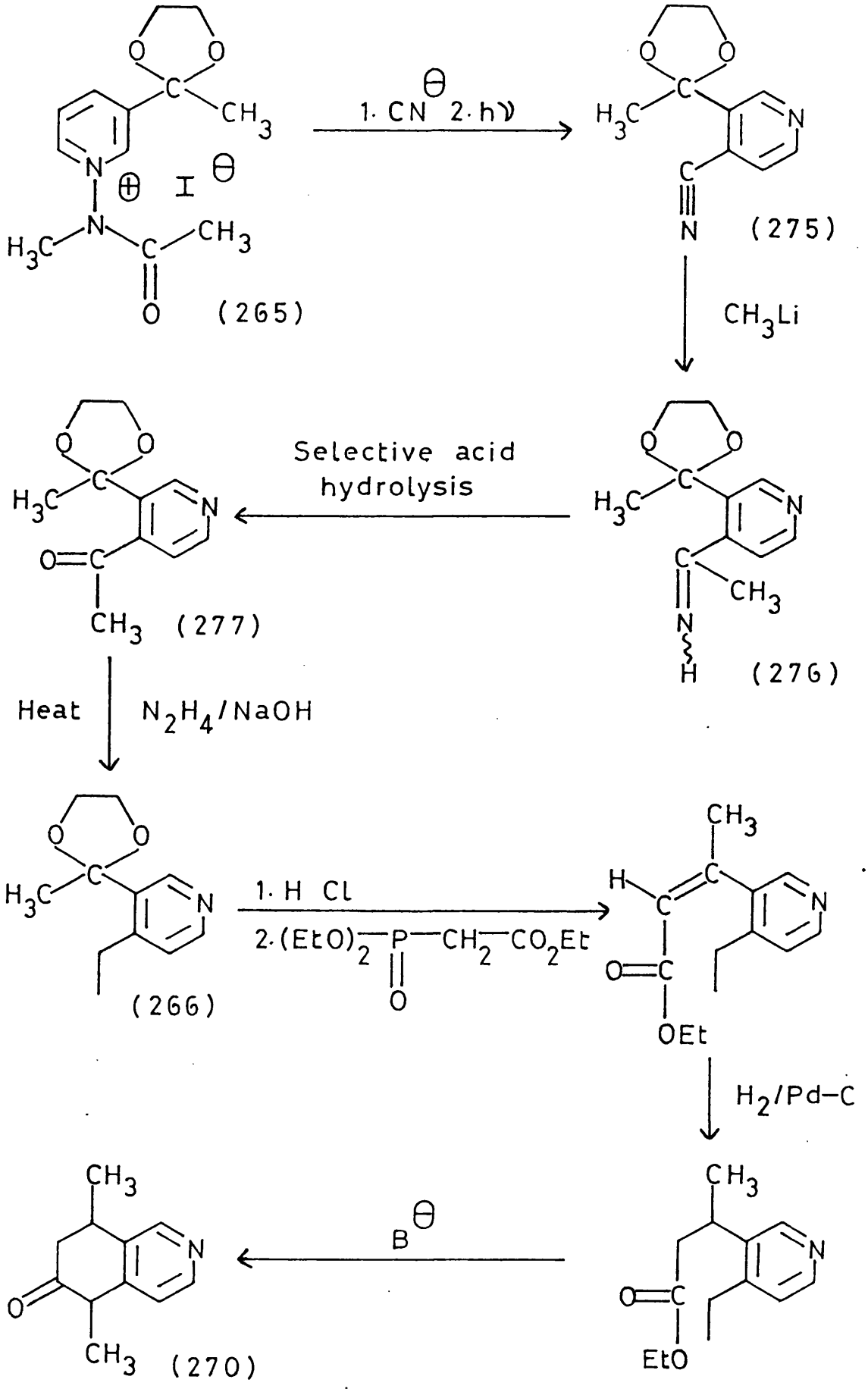
(274)

As Katritzky and Sammes technique for aromatizing such dihydropyridine intermediates is to heat them at temperatures in excess of 200°, rather than irradiating, we repeated our experiment, but heated the residue from the Grignard reaction at 200-250° for four hours in the hope of causing a similar rearrangement to occur. However, on working up the reaction in the usual manner, the only product isolated was the 4-ethylpyridine (273).

Thus, we concluded that if such rearrangements do occur they are substituent dependent and are not successful with alkyl groups.

Due to this unexpected result we were unable to implement the remainder of the synthesis previously outlined in (Scheme 77), but a possible alternative is that shown in (Scheme 78).

Scheme 78.


1. CN⊖ 2. hν
(265)
(275)
CH3Li
Selective acid hydrolysis
(277)
(276)
Heat
N2H4/NaOH
(266)
1. H Cl
2. (EtO)2P(O)CH2CO2Et
OEt
H2/Pd-C
B⊖
(270)
EtO

It has been known for some time in our laboratory[18] that the small, nucleophilic cyanide ion is capable of substituting α- to the bulky cyclic acetal group of the pyridinium methiodide (265). We carried out this reaction by treating an aqueous solution of (265), with potassium cyanide in the presence of ammonium chloride at room temperature, after extraction of the 1,4-dihydropyridine intermediate (not shown), and irradiation of an ethanolic solution of this with ultraviolet light, a good yield, 87% of the carbonitrile (275) was obtained.

Lack of time prevented further work on this promising approach by the author, but exposure of (275) to methyl lithium would give the methyl imine (276). Selective acid hydrolysis would then afford the pyridyl ketone (277). Wolff-Kishner reduction of the carbonyl group with hydrazine hydrate in sodium hydroxide solution may then give the 4-ethylpyridine (266), as before. This might then be converted to the key cyclic ketone (270) and hence to 8-hydroxyellipticine as outlined previously in (Scheme 77, p. 212).

It is hoped that further work in the future will clarify the potential of this new synthetic approach.

PHARMACOLOGY

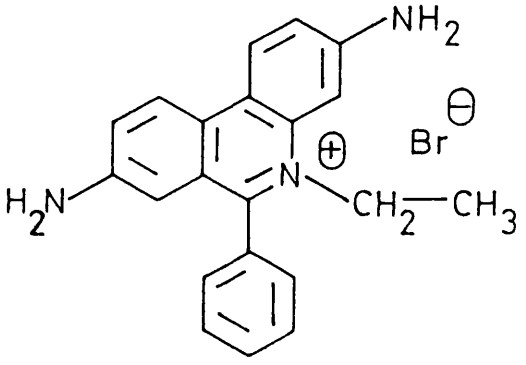
Ellipticine and its derivatives as chemotherapeutic agents

Following the initial isolation of ellipticine and 9-methoxyellipticine,[1] these alkaloids were evaluated for biological activity. Initially, this proved disappointing because high dose rates of ellipticine were found to cause respiratory failure in mice[199]. However, the next year (1968) Svoboda and his co-workers[200] investigated ellipticine and 9-methoxyellipticine as possible antineoplastic agents. These workers found that both compounds were active against tumour cells, but 9-methoxyellipticine was shown to be particularly effective against various mouse experimental tumour systems, so that lower dose rates than before could be employed. The Walker 256 rat carcinosarcoma was also seen to respond, when this compound was administered. Particularly encouraging was the work of Le Men et al[201] who demonstrated that the water soluble lactate of 9-methoxyellipticine inhibited mouse L1210 III murine leukemia. This neoplasm is resistent to many established anti-cancer drugs, and many agents that are active against this tumour (which closely resembles the human situation), have been found to be effective against cancer in man. Hill and Baserga[202] have also shown this compound to have a cytostatic effect on many types of leukemic cells and preclinical trials with dogs showed a low toxicity and good tolerance to the drug. Human clinical trials were initiated in France by Mathe[68], who has shown that this compound has significant activity against acute myeloblastic leukemia, and out of twelve patients treated three complete remissions were reported.

It is now generally accepted[57 and 203] that the mode of action of many cytotoxic agents such as the ellipticines or the acridines[204] against cancerous cells is due to intercalation between adjacent layers of base pairs in D.N.A.[205] with subsequent inhibition of cell replication. The underlying reasons for the selective toxicity of these drugs against cancerous cells compared with normal cells is not well understood. However, it appears[206] that in some tumour cells the normal molecular repair mechanisms are less effective than in normal cells and hence damage by intercalating agents has a greater effect. The size and shape of such drugs is obviously critical, as for the drug to be effective it must be able to fit into the cavities of the double helical structure of D.N.A., and bind to the adjacent layers of base pairs.

The normal process of D.N.A. replication involves the unwinding of the double stranded structure and the formation of new chains which are complementary to each strand. The hydrogen bonded base pairs Adenine-Thymine and Cytosine-Guanine have been found to be the only stable ones for D.N.A. construction[207], (see Fig. 11). The thermodynamics of the situation dictate that only a few bases unwind at a time, as ($\sim$ 240 K Joules/mole) are required to separate the components of a single pair of bases. Thus all the bases will pass through the single stranded condition, but not at once; in this way a disturbance is propagated along the whole D.N.A. chain, and much of the energy required for base separation is then supplied by bases reuniting. Hence it is clear that this process will require a perfect D.N.A.

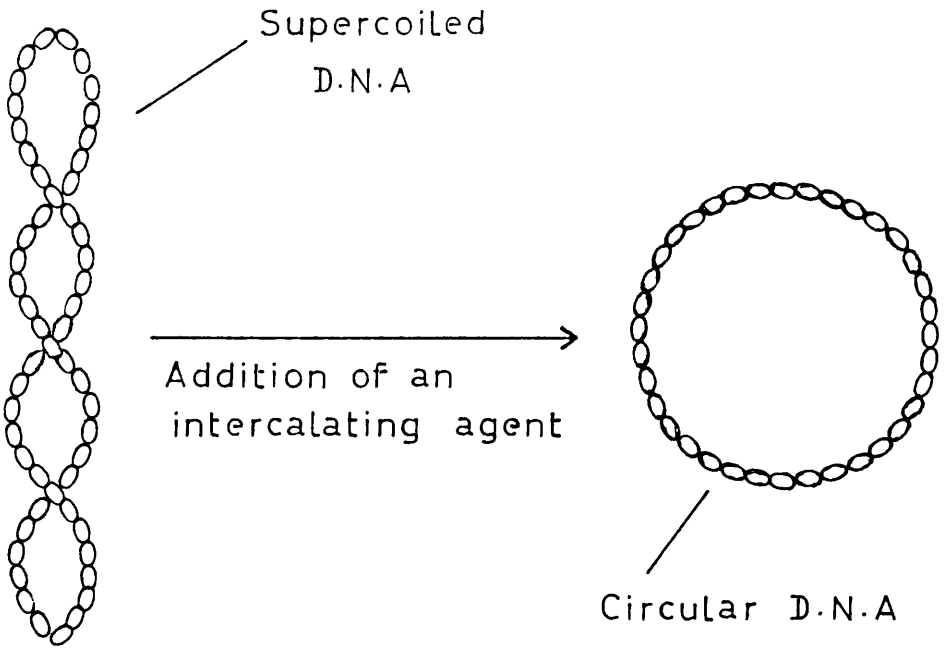
molecule to be effective. When molecules of an intercalating drug are inserted into cellular D.N.A. distortions occur in the helical structure at the points where these molecules are bound across the chain (see Fig. 11). L. Lerman[208] has shown that when intercalating molecules are taken up by a D.N.A. helix, spaces comparable in thickness to a nucleotide base pair have to be provided for them between the existing base pairs. This is achieved by the chain unwinding slightly at these points to admit the intercalating molecules. This unwinding angle is variable, homidium bromide[209] (278), gives a value of 26° and for the ellipticine alkaloids it is rather less (see Table 5).



(278)

Some D.N.A. molecules particularly in bacteria[210] have a circular unbroken D.N.A. chain, and in some cases the helical chain is itself coiled, so as to produce super coils[211]. An interesting effect of introducing intercalating molecules into the helix of this type of D.N.A. is that the super coils unwind giving simple open circular D.N.A.[212]. (see Fig. 10).

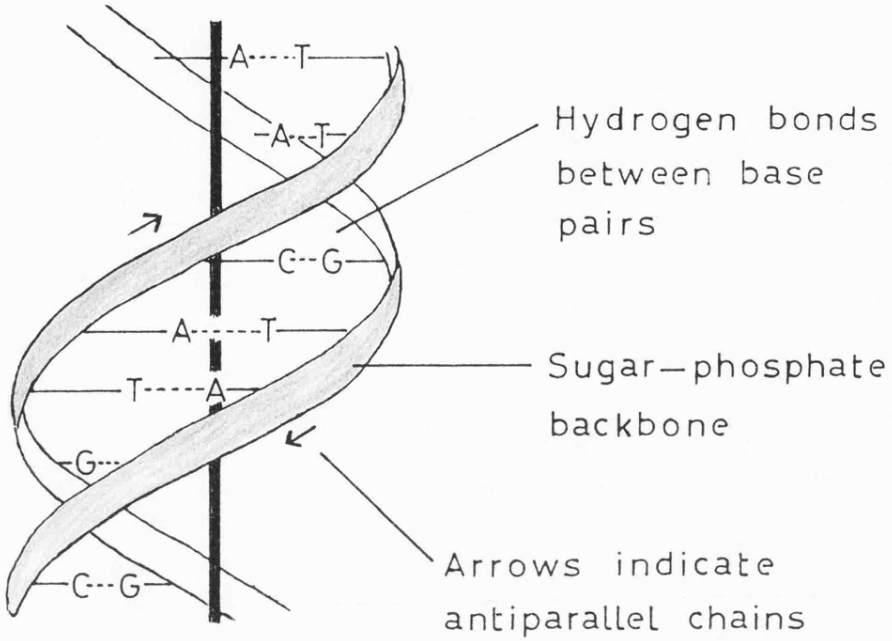
Fig. 10



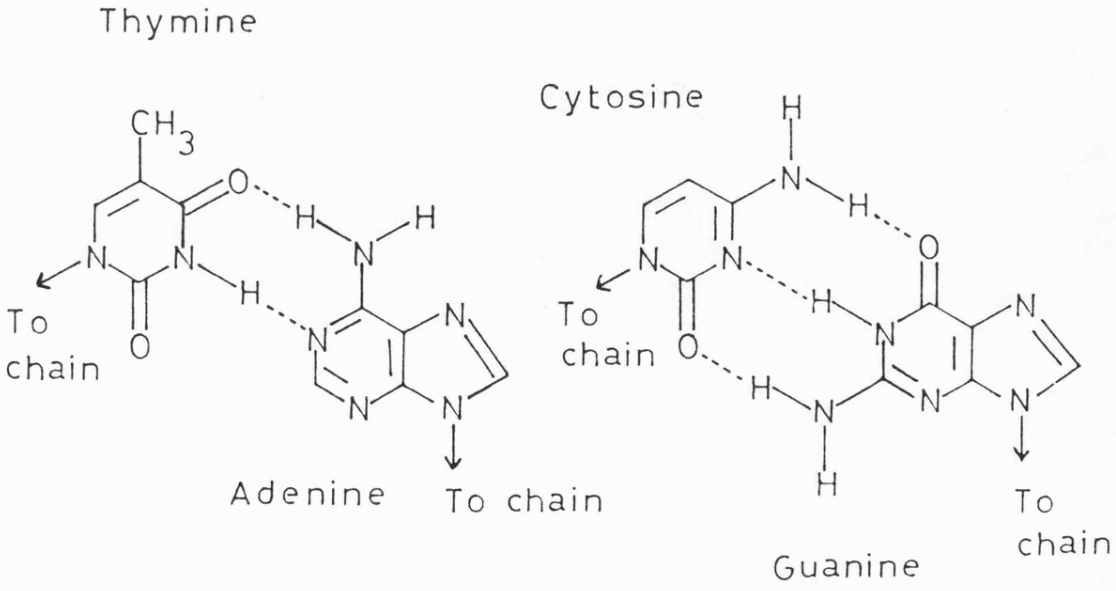
The circular D.N.A. is of greater perimeter than normal due to the stretching effect of the intercalated molecules. If even more of the intercalating agent is added the stretching reaches a point so great that the D.N.A. will form a distorted super coil again, but this time in the opposite direction to the original (right handed $\rightleftharpoons$ left handed). As super coils are more compact than the open circles the whole process is easily monitored by changes in the sedimentation coefficient which passes through a minimum.

Fig. 11.

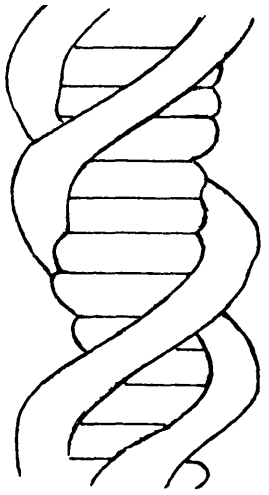
Schematic diagram of the D.N.A. double helix



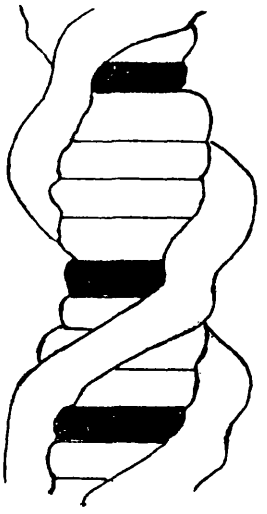
Hydrogen bonded base pairs in D.N.A.



Schematic projection of D.N.A. distortion caused by intercalating molecules.



Normal D.N.A



Distorted D.N.A

The intercalation of such molecules causes other changes to occur in D.N.A. When proflavin[213] was intercalated with D.N.A. it was found that the helix was extended and stiffened so that a threefold increase in viscosity took place. It can be seen from these facts that such gross changes in the structure of a D.N.A. molecule will inhibit its replication with subsequent cell death.

Two factors are important in the structure of an intercalating molecule. Firstly the electronic configuration of the molecule decides the magnitude of the interaction energy. That is, the strength of the hydrogen bonds, Van der Waals forces,

hydrophobic and hydrophilic interactions, all of which go to make up the total binding with D.N.A. Secondly stereochemical parameters control the ability of a molecule to intercalate, and planarity or a close approach to it is essential for this effect. The flat, arc-shaped 6H-pyrido[4,3-b] carbazoles have this property as do the acridines. French workers[203] have shown that 9-methoxyellipticine intercalates with D.N.A. at high and low ionic strengths. This effect is obviously reduced when bulky groups are introduced around the periphery of the ring system. Direct evidence in support of this has been obtained in this laboratory[18], when 9-phenylellipticine was found to be inactive because the phenyl substituent is twisted out of plane with respect to the rest of the tetracyclic skeleton, preventing overall planarity of the molecule and hence intercalation. In general any bulky group will inhibit activity and possibly the lack of success with isopentylellipticine (see Table 5), can also be attributed to this. It is possible that very large atoms substituted directly into the ring system have a similar effect, as 9-bromoellipticine is unable to intercalate and hence is inactive.

Of the many ellipticine derivatives synthesized in recent years, Mathe et al[68] have shown that 9-hydroxyellipticine has the highest degree of antineoplastic activity of any known ellipticine (see Table 5). Further development of this work by Le Pecq, Dat-Xuong, Paoletti and Gosse[214] has seen an attempt to rationalize the approach to ellipticine design, so as to maximize the structural features required for significant

activity. These workers measured some physiochemical properties for a series of ellipticine derivatives and tried to correlate their results with the observed anti-cancer activity. They consider that for intercalating drugs such as ellipticine and its derivatives a high D.N.A. binding efficiency is a necessary condition for anti-neoplastic activity.

Ellipticine can exist in a protonated and a neutral form and the D.N.A. binding affinity of the cationic form of the drug KE^{+} is about 30 times larger than the D.N.A. affinity of the neutral form. In aqueous solution under physiological conditions at pH 7.4 both protonated and neutral forms are present and the apparent binding constant then measured is dependent upon the pKa. The D.N.A. binding constants were determined from the following equation

$$\log Kap = \log KE^{+} - \log \left[1 + \frac{KH^{-1}}{[H^{+}]} + \log \left(1 - \frac{1}{\alpha} \frac{KH^{-1}}{[H^{+}]} \right) \right]$$

where Kap = D.N.A. binding constant of the derivative

KE^{+} = D.N.A. binding constant for the protonated form

KH^{-1} = Dissociation constant of the equilibrium between the protonated and non-protonated forms of the derivative.

α = The ratio of the D.N.A. binding constants of the protonated and neutral forms of the drug.

Thus it would appear that the pKa is one of the main factors affecting D.N.A. affinity at physiological pH. The pharmacological activity was expressed as a percentage of the L1210 mouse leukemia cells killed by 1/3 of the dose that would prove lethal to 50% of the animals, or the (LD_{50}) value. The D.N.A. unwinding angle was also measured for a number of ellipticine derivatives and the results are shown in(Table 5.)

Table 5.

Substituent	pK	Kap (pH 7.4)	Log KE^+	Unwinding angle.	% of cells killed at 1/3 LD_{50}
6-isopentyl	4.7	$1.0x10^4$	6.3	8.8	0
5,11-didemethyl	6.35	$1.0x10^4$	5.1	-	-
11-demethyl	6.3	$2.4x10^4$	5.5	-	0
9-methoxy	6.8	$1.0x10^4$	5.7	6.8	90
ellipticine	9.1	$1.5x10^4$	5.2	9	94
9-bromo	6.1	$4.0x10^5$	6.9	0*	0
6-methyl	6.1	$4.0x10^5$	6.9	10.2	92
9-amino	9.8	$1.2x10^6$	6.1	4	0
9-hydroxy	9.8	$2.0x10^6$	6.2	12	99.96
9-methoxy-6-methyl	6.45	$2.0x10^6$	7.3	5	50

* 9-Bromoellipticine does not intercalate.

From the table it can be seen that there is a partial correlation between the magnitude of the D.N.A. unwinding angle and pharmacological activity. In general the larger the unwinding angle the greater the number of cells killed,

presumably because the molecules giving rise to these effects cause the greatest degree of D.N.A. distortion and hence are most effective in promoting the unwinding process, and thus inhibiting replication. However, a number of factors must be considered when interpreting these results. Firstly, as mentioned previously (see p. 227), stereochemical constraints are important, and bulky substituents can inhibit or entirely prevent intercalation. Secondly the unwinding angle is measured in vitro with cell free D.N.A. molecules[215], but the ($1/3\ LD_{50}$) values are obtained by working with whole cell preparations[216]. Hence, cell permability may play a part in determining the overall effect of any particular derivative. It is not possible, therefore, to draw any definite conclusions about apparent inconsistances in this type of data.

It is interesting to note the 3-unit drop in pK value and the consequent decrease in D.N.A. binding constant when the methyl group at C-11 is replaced by a hydrogen atom. One possible explanation for this is that the electron donating ability of the C-11 methyl group is contributing to the electron density at the pyridine nitrogen, hence its removal lowers the basicity of this nitrogen and therefore the D.N.A. binding constant. It is also believed that the C-5 and C-11 methyl groups confer a degree of lipid solubility on the aromatic skeleton, which increases the ease of transport across cell membranes. Removal of one or both of the methyl groups may cause partial, or total loss of this property, and physically prevent the demethylellipticine molecules from reaching the D.N.A. in the interior of the cell.

Interest in the synthesis of ellipticine derivatives has centred around attempts to produce structures with a high level and wide spectrum of anti-cancer activity, and hence, the hoped for elucidation of structure activity relationships. Unfortunately it is not possible to decide intuitively the best derivatives to prepare. However, by analysing a combination of electronic, steric and solubility characteristics, Hantsch[217], in 1972 arrived at a theory which purports to indicate the best choice of synthetic targets. Shortly after this a number of ellipticines were synthesized by chemists all over the world, but it is clear from (Table 6) that most of the pyrido[4,3-b]carbazoles synthesized at this time reflect their ease of access rather than a determined attempt to prepare those with structural elements most likely to reveal biological activity. However, more recent work in this laboratory[218,219] and elsewhere have produced several new synthetic ellipticines with the 'A' or 'C' rings bearing a range of substituents (see Table 7). These compounds are undergoing pharmacological evaluation and the results are awaited with interest.

Table 6

Substituents in Derivative	m.p. °C	Reference
1,2-dihydro	281-283(dec)	2
1,2,3,4-tetrahydro	163-165(dec)	15
2-methyl-1,2,3,4-tetrahydro	302-307(dec)	2
6-methyl		223
9-methyl		223
3-methyl		223
9,10-dimethyl		223
6-isopentyl		
9-hydroxy	330	57
9-acetoxy	282	57
9-acetoxy-6-methyl	185	57
9-methoxy	286-287	12
9-methoxy-6-methyl	315	57
9-amino	255-260	57, 17
9-bromo	318-319	12
9-nitro	350-357(dec)	12
9-phenyl	307-308	18
6-acetyl-9-methoxy	159-160	170
9-ethoxycarbonyl	280-281(vac)	170
9-methoxycarbonyl	303-304(vac)	170
9-n-pentoxycarbonyl	235-236	170
6-methyl-9-n-pentoxycarbonyl	151-152	170
9-phenoxycarbonyl	294-296	170
9-n-dodecanoxycarbonyl	189-190.5	170
9-n-heptadecanoxycarbonyl	183-185	170
9-(1-admantanoxycarbonyl)	350	170
9-n-butoxy	248-249(vac)	170

Substituents in Derivative	m.p. °C	Reference
8,9-methylenedioxy	330-333(dec,vac)	170, 18
8,9-dimethoxy	315-319(dec,vac)	170
9-hydroxy-6-methyl	315	57

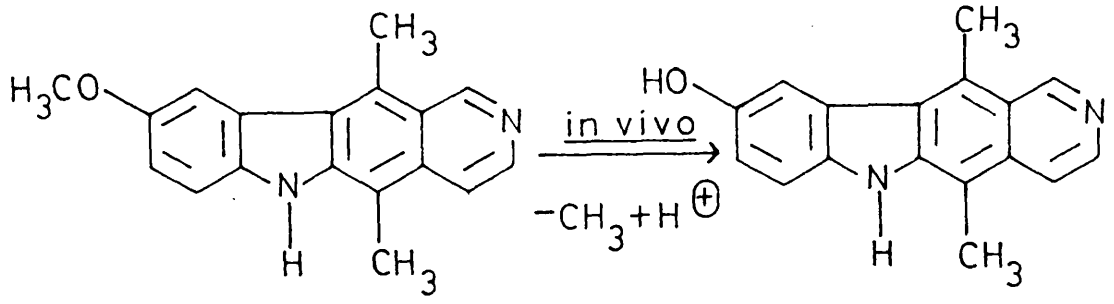
Where dec = decomposition

and vac = determined under vacuum.

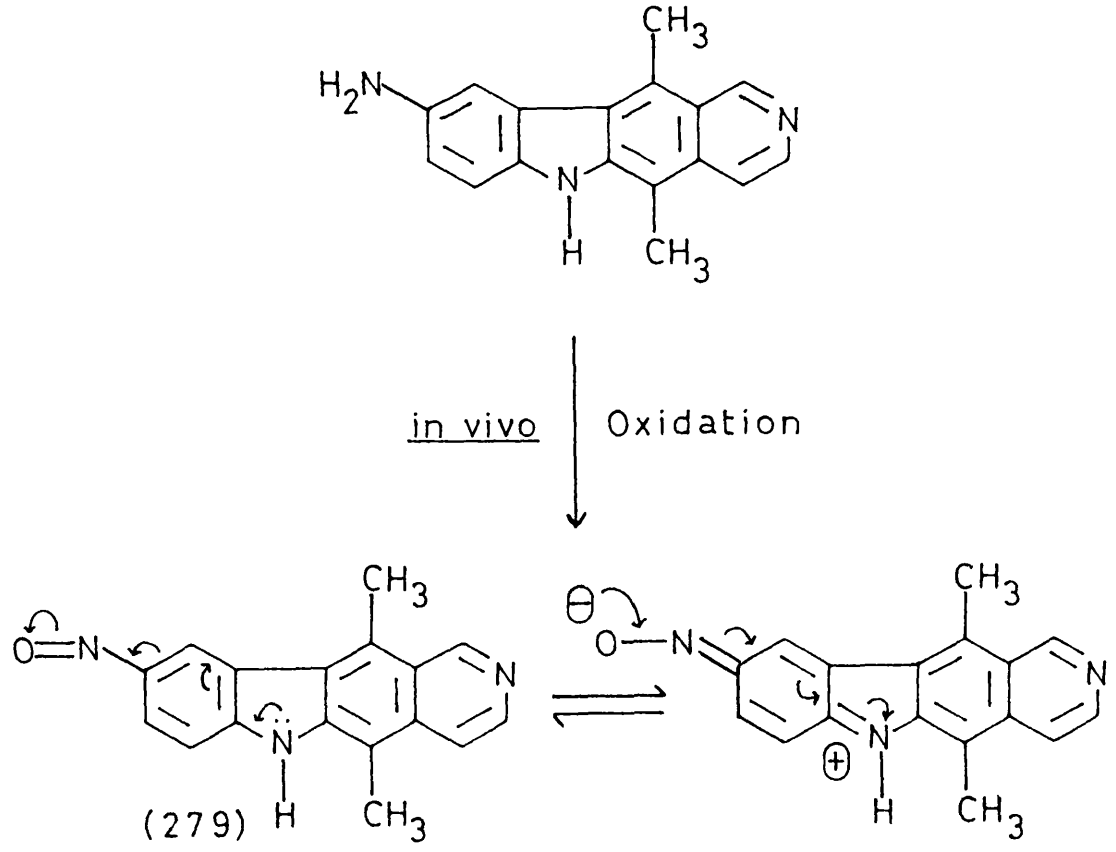
Table 7.

Substituents in Derivative	m.p. °C	Reference
8-methoxy	280-281	218
8-Hydroxy	268-270	218
7-Hydroxy	-	56
5-n-Butyl-11-methyl	285-287	69
6H-Benzo h pyrido [4,3-b] carbazole	327-329	174
7-Fluoro	244-245	219
7-Chloro	315-320	219
7-Methyl	298-300	219

Since the discovery of the high level anti-neoplastic activity of 9-hydroxyellipticine it has been suggested[220], that the good results observed with 9-methoxyellipticine are due to metabolic O-demethylation of this compound, followed by protonation to give 9-hydroxyellipticine.

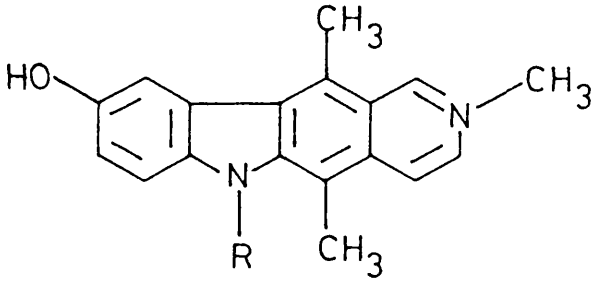


Another interesting fact emerged when French workers[57] studied the metabolism of 9-aminoellipticine. This compound proved to be inactive against experimental tumour systems, and surprisingly exhibited carcinogenic properties. This is now considered to be due to biological oxidation of this compound to give nitroso compounds or intermediates such as (279).



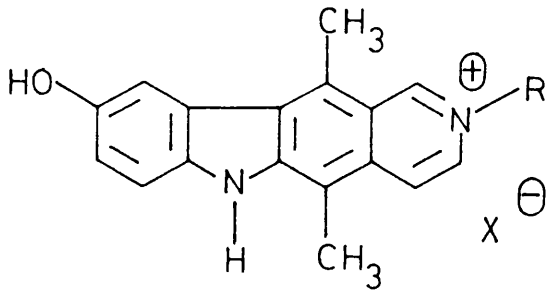
(279)

From (Table 5) it is apparent that 9-hydroxyellipticine is the most potent anti-neoplastic agent among the ellipticine derivatives that have been biologically tested to date. This derivative kills more than 99% of the mouse L1210 leukemia cells at 1/3 LD_{50}. This naturally lead to further work with hydroxy ellipticines, including our own synthesis of 8-hydroxyellipticine. The 2-methyl and 2,6-dimethyl analogues of 9-hydroxyellipticine (280) and (281) have also been synthesized and appear to be even more active.[214]



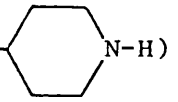
(280, R = H and 281, R = CH_3)

As these compounds are also salts, they are water soluble, which is an obvious clinical advantage as they are easier to administer to the patient. This approach has been extended[221] to other 2-substituted 9-hydroxyellipticines (282, 283, 284, and 285), which are undergoing pharmacological evaluation.



(282, R = CH_2CH_2OH), (283, R = CH_2CH_3)

(284, R = $CH_2CH_2N(Et)_2$)

(285, R = CH_2CH_2 )

As only a relatively small number of ellipticine derivatives have been biologically tested the wide spectrum and level of anti-cancer activity is most encouraging. The main problem with the clinical evaluation of these alkaloids has been one of supply, as they occur in low abundance in the tropical plant species that synthesize them. Therefore it is not surprising that so much effort has been devoted to obtaining a satisfactory synthetic route[222] as outlined in the introductory section (p. 3 - 64), and some interest has been generated in the recent work involving microbial hydroxylation of the ellipticine skeleton. Future developments of the latter, may prove fruitful as ellipticine is a little easier to synthesize than its hydroxy

analogues. However, it is hoped that the work described in this thesis will promote further interesting investigations and provide a means of synthesizing 8-hydroxyellipticine on a larger scale, to allow the full pharmacological evaluation of this new derivative.

EXPERIMENTAL

Ultraviolet spectra were recorded on a Perkin-Elmer 402 Ultraviolet and Visible Spectrophotometer in 95% aqueous ethanol. Infrared spectra were obtained with a Perkin-Elmer 137 grating Spectrophotometer as Nujol or hexachloro-1,3-butadiene mulls, liquid films or as 5% chloroform solutions. Other abbreviations used are st = stretch and def = deformation.

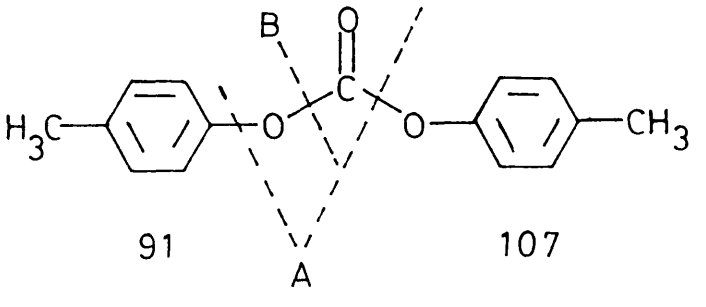
^{1}H n.m.r. spectra were run on JEOL instruments either at 100 MHz and 1080 Hz sweep width or, for high resolution studies, at 200 MHz with a 4000 Hz sweep width on a Fourier transform (F.T.) instrument. The chemical shift data are expressed as δ values with respect to tetramethylsilane (T.M.S.) as internal standard. Other abbreviations used are: s = singlet, d = doublet d x 2 or d x d = double doublet, t = triplet, q = quartet and m = multiplet.

Mass spectra were recorded on a spectrometer comprising a A.E.I. MS12 source coupled to a Vg micromass recorder, run at 70eV for routine measurements and at 12eV for molecular ion detection

p-Tolylcarbonate (135)

To a rapidly stirring solution of 4-cresol (129 g ≡ 1.2 mol) and sodium hydroxide (50 g ≡ 1.25 mol) in water (600 cm^3) was added phosgene in toluene (500 cm^3 of a 12.5% solution ≡ 60 g of $COCl_2$ or 0.63 mol). The addition was carried out portionwise over a period of thirty minutes, maintaining the temperature at 40-50^o by means of a thermostatic bath. The mixture was stirred

at this temperature for three hours, allowed to cool to room temperature and the stirring continued for a further eighteen hours. Nitrogen gas was then bubbled through the solution for thirty minutes to free it of residual phosgene, the aqueous layer was separated and extracted with dichloromethane (3 x 100 cm^3). The toluene and dichloromethane layers were combined and thoroughly washed with cold water (4 x 100 cm^3), dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a white solid. Recrystallization from toluene gave fine white needle crystals (90 g, 62%), m.p. 112-113°. (lit[92]; 112-113°). $\lambda_{max}(\varepsilon)$ 254 (16,500), 335 (4,120), 340 (4,050), 359 (3,500)nm. ν_{max} 1760 (C=O st), 1600 (Ar C-H st), 1030, 980, 810 and 750 cm^{-1}. δ ($CDCl_3$) 2.35 (6H, s, 2 x $\underline{CH}_3$), 7.30 (8H, s, aromatic $\underline{H}^s$). The simplicity of this spectrum illustrates the completely symmetrical nature of this molecule. m/e 242 (100%, base and $M^{+\cdot}$), 198 (89), 184 (8), 171 (7), 156 (22), 108 (24), 91 (98), 78 (23). This spectrum exhibited a fine set of meta stable ions, from which it is apparent that the molecule has two alternative fragmentation pathways. A. Loss of CO_2, B loss of CO. Metastables as follows: 163.64 for 242 - 198 - 44 CO_2, 146.94 for 198 - 171 - 28 CO, 142.32 for 171 - 156 - 15 CH_3, 45.00 for 184 - 91 - 93 CH_3-C_6H_4, 41.60 for 199 - 91 - 107 CH_3-C_6H_4O.



4-Methyl-3-nitrophenol (138, Method A)

A solution of p-tolylcarbonate (72 g = 0.297 mol) in concentrated sulphuric acid (360 g = 196 cm^3) was prepared by careful portionwise addition of the carbonate to the rapidly stirring acid maintained at less than 10^o by means of an ice-salt bath.

When addition was complete the solution was cooled to 0^o and a ice-cold mixture of concentrated sulphuric acid (72 g = 39 cm^3) and concentrated nitric acid (40 g - 30 cm^3) added dropwise with continuous stirring, maintaining the temperature between 5 - 10^o. When addition was complete the mixture was allowed to warm to room temperature and the stirring continued for a further two hours. The mixture was then poured onto crushed-ice (600 g).

This gave a dark orange precipitate. Analysis of this product by T.L.C., G.L.C., I.R and mass spectral techniques showed it to consist of a mixture of the 2 and 3-mononitro-4-tolylcarbonates. The product ratio was 3:2 for the desired 3-nitro-4-tolylcarbonate and the 2-nitro-4-tolylcarbonate respectively.

The mixture of mononitrotolylcarbonates was redissolved in 10% aqueous sodium carbonate solution (600 cm^3). The alkaline mixture was stirred and heated at reflux temperature for two hours, to hydrolysis the nitrocarbonates, cooled to toom temperature and acidified with concentrated hydrochloric acid to liberate the free phenols.

The reaction mixture was then steam distilled to remove 4-methyl-2-nitrophenol*. However, the residual 4-methyl-3-nitrophenol was still too impure to be isolated directly, so it was extracted with chloroform (4 x 200 cm^3). The organic extracts were combined and dried over annydrous sodium sulphate, the drying agent was removed and the filtrate treated with decolourising carbon at reflux temperature for five minutes. The carbon was filtered off from the hot solution and the chloroform evaporated under reduced pressure to give a red gum. This material was chromatographed on silica gel, eluting with chloroform. The first yellow band emerging from the column was collected and evaporated under reduced pressure to give an amorphous, amber coloured solid. This nearly pure material was crystallized from 95% aqueous ethanol to give pure 4-methyl-3-nitrophenol as shining amber plates (36.4 g, 40%), m.p. 79-80°. (lit[92] 76-77°), $\lambda_{max}(\varepsilon)$ 242 (12,180), 271 (6,410), 344 (3,525) nm. ν_{max} 3,400 (broad, H-bonded -OH), 1600 (Ar), 1510 (NO_2 antisymmetric stretch), 1200(O-H deformation), 840 (C-N stretch), 880 and 810 (1,2,4-aromatic substitution pattern), 760/740 (doublet C-N in plane deformation)cm^{-1}. δ ($CDCl_3$) 2.50 (3H, s, $C\underline{H}_3$), 5.6 (1H, s broad, O$\underline{H}$, exchanges with D_2O), 7.05 (1H, d x 2 $J_{5,6}$ 7Hz, $J_{5,3}$ 2Hz - 5$\underline{H}$), 7.2 (1H, d $J_{6,5}$ 7Hz - 6$\underline{H}$), 7.55 (1H, d $J_{3,5}$ 2Hz - 3$\underline{H}$). m/e 153 ($M^{+\cdot}$ 64%), 136 (100 base), 108 (43), 78 (60). Metastables at 120.8 corresponding to 153 - 136 - OH), and 85.76 for 136 - 108 corresponding to -CO.

4-Methyl-2-nitrophenol* (139)

The steam distillate from the foregoing preparation of 4-methyl-3-nitrophenol was extracted with dichloromethane (4 x 200 cm^3). The organic extracts were combined, dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a yellow amorphous solid. This material was crystallized from 95% aqueous ethanol over a period of three days at -20° to give pure 4-methyl-2-nitrophenol as shining, bright yellow needles (23.8 g, 26.1%) m.p. 32-34°. (lit.[224] m.p. 36°), λ_{max} (ε) 242 (11,600), 278 (6,400), 351 (3,500)nm. ν_{max} 3,500-3,200 (H-bonded O-H st), 1600 (Ar-H st), 1510 (NO_2 antisymmetric st), 1330, 1200 (O-H def), 1100, 910, 830 (C-N st), 870 and 800 (1,2,4-aromatic substitution pattern), 780 and 760 (doublet C-N in plane def) cm^{-1}. δ ($CDCl_3$) 2.35 (1H, s, $\underline{CH}_3$) 7.0 (1H, d $\underline{J}_{5,6}$ 8Hz-5$\underline{H}$), 7.4 (1H, d x 2 $\underline{J}_{6,5}$ 8Hz, $\underline{J}_{6,2}$ 2Hz - 6$\underline{H}$), 7.8 (1H, d $\underline{J}_{2,6}$ 2Hz - 2$\underline{H}$). The O-H proton signal does not appear in the normal range of 0 - 10.8 p.p.m., presumably because this isomer exists as an intramolecularly hydrogen bonded chelate. m/e 153 $M^{+\cdot}$

p-Toluene ethylcarbonate (140).

4-Cresol (150 g = 0.72 mol) was dissolved in dry pyridine (200 cm^3) and ethylchloroformate (150 g = 0.72 mol) added dropwise over a period of one hour with continuous stirring, maintaining the temperature at 0 - 10° by means of an ice-salt bath. When addition was complete the solution was allowed to warm to room temperature and the stirring continued for a further hour.

The reaction mixture was then poured into ice-water (600 cm^3) and extracted with ether (3 x 200 cm^3). The ether extracts were combined and washed with 2N hydrochloric acid (4 x 300 cm^3). The combined organic phases were then dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a colourless liquid. This product was distilled at 108°/0.3 mm pressure to give pure para-toluene ethylcarbonate as a colourless oily liquid (240 g, 96%). λ_{max} (ε) 252 (11,200), 340 (3,200), 335 (3,150), 352 (2,500) nm. ν_{max} 1760 (C=O st), 1260 and 1220 (ø-C-O st), 1200 (C-O-Et st), 1050, 1000, 810 (1,4-aromatic substitution pattern)cm^{-1}. δ ($CDCl_3$) 1.3 (3H, t, CH_3CH_2), 2.3 (3H, s, Ar-CH_3), 4.25 (2H, q, CH_3CH_2), 7.15 (4H, m, aromatic H^s). m/e 180 ($M^{+\cdot}$, 100% base).

Nitration of p-toluene ethylcarbonate

Preparation of 4-methyl-3-nitrophenol (Method B).

p-Toluene ethylcarbonate (107 g = 0.594 mol) was nitrated exactly as described in (Method A, p 240). However, the reaction proved to be more efficient, and cleaner than the results obtained with p-tolylcarbonate. The steam distillation residue could be crystallized directly from ethanol to give 4-methyl-3-nitrophenol as orange plates. The yield was (61 g, 67%) compared with 40% for p-tolylcarbonate. The physical data for this product were identical in every respect with those quoted on p . 241 .

4-Benzyloxy-2-nitrotoluene (141).

To a solution of 4-methyl-3-nitrophenol (18.6 g = 0.12 mol) in 35% ethanolic potassium hydroxide (170 cm^3) was added water (40 cm^3) and benzyl chloride (30 g = 0.24 mol). The dark red solution was heated at reflux temperature for three hours, cooled to room temperature and the alcohol removed by evaporation under reduced pressure.

The residue was extracted with chloroform (4 x 150 cm^3). The combined organic layers were washed with 10% aqueous sodium carbonate solution (2 x 100 cm^3), N-sodium hydroxide solution (2 x 100 cm^3) followed by water (2 x 100 cm^3). The chloroform layer was dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a clear red oil. Trituration with petroleum ether (b.p. 40 - 60°) yielded an amorphous orange solid. This material was crystallized from a 1:1 mixture of diethylether/petroleum ether (b.p. 40 - 60°) to afford pure 4-benzyloxy-2-nitrotoluene as pale yellow plates (25.7 g, 87.2%) m.p. 51 - 52° (lit;[89] m.p. 52°) λ_{max} (ε) 243 (3,600), 265 (2,500), 339 (1,200) nm. ν_{max} 1600 (Ar), 1540 (NO_2 antisymmetric stretch), 1360 (NO_2 symmetric stretch), 1240 (C-O-C aralkyl stretch), 110, 1020, 860 (C-N stretch), 840 and 790 (1,2,4-aromatic substitution pattern),750 (C-N out of plane deformation)cm^{-1}. δ ($CDCl_3$) 2.50 (3H, s, $\underline{CH}_3$), 5.05 (2H, s, $\underline{CH}_2$), 7.10 (1H, d x 2 $\underline{J}_{5,6}$ 7Hz $\underline{J}_{5,3}$ 2Hz - 5$\underline{H}$), 7.20(1H, d $\underline{J}_{6,5}$ 7Hz - 6$\underline{H}$), 7.3-7.5 (5H, m, Benzyl ring $\underline{H}^s$), 7.60 (1H, d, $\underline{J}_{3,5}$ 2Hz - 3$\underline{H}$).

m/e 244 (M + 1 5%), 243 ($M^{+\cdot}$ 20), 213 (4), 198 (12), 152 (3), 136 (5), 107 (11), 91(100 base), 77 (9), 46 (90). The two most significant peaks in the spectrum apart from the molecular ion occur at m/e values of 91 and 46, which correspond to the simple fission of the benzyl and nitro groups respectively.

4-Benzyloxy-2-nitrophenylethylpyruvate (142)

Sodium dried xylene (200 cm^3) was placed in a three necked, round bottom flask (1 l. capacity), fitted with a dropping funnel (250 cm^3 capacity), nitrogen inlet, motor driven stirrer with a glass paddle and a sealing gland and a reflux condensor protected with a calcium chloride guard tube.

Potassium (freshly cut into small pieces and degreased with petroleum ether (b.p. 40 - 60°) under a stream of dry nitrogen) (4.08 g = 0.104 g atom), was added quickly against a positive nitrogen pressure. A slow stream of dry nitrogen was passed through the flask, above the surface of the stirred liquid, and the mixture was slowly raised in temperature, with constant stirring until the potassium was seen to melt. The temperature was held at 60° for 10 minutes and then allowed to cool to room temperature with constant stirring, to give finely powdered potassium as a grey solid.

When the solution reached room temperature, a mixture of absolute ethanol (4.75 cm^3) and ether (15 cm^3) was added dropwise at such a rate as to maintain a gentle reaction. When all the potassium had dissolved, approximately 4 hours, the nitrogen was

shut off and the solution allowed to cool to room temperature. The solvents were evaporated under reduced pressure to give potassium ethoxide (8.83 g = 0.105 mol = 1.03 mol equivalents) as a white powder.

The potassium ethoxide was dispersed in diethylether (200 cm^3) and freshly distilled diethyl oxalate (16.0 g = 15.0 cm^3 = 0.109 mol or 1.03 mol equivalents) was added dropwise with stirring. The mixture was cooled to 0 - 5^o in an ice-salt bath and 4-benzyloxy-2-nitrotoluene (25.0 g = 0.102 mol) was added portionwise with stirring. The reaction was decidedly exothermic and the colour changed from pale yellow to dark amber. The mixture was then heated at reflux temperature for 16 hours and allowed to cool to room temperature. The dark red solution was poured into ice-water (500 cm^3), whereupon a sticky yellow solid separated. This solid was too oily to allow filtration so it was extracted with dichloromethane (4 x 75 cm^3), the combined organic phases were dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a red oil. Trituration with ether gave an orange solid. This solid was crystallized from ethanol to give the pure product as a light amber coloured solid. (19.03 g , 54%) m.p. 88-89^o λ max (ε) 215 (14,100), 236 (7,000), 268 (3,600) and 348 (1,300) nm. ν max 1765 (O=C-OEt st), 1740 (C=O st), 1600 (Ar-H st), 1520 (Ar-NO_2 st), 1340 (C-N st), 1150 (C-O st), 1090 and 1010 (C-O-C st), 840 (C-N def), 900 and 800 (1,2,4-aromatic substitution pattern) 750 and 700 (aromatic, mono substitution pattern) cm^{-1}.

δ ($CDCl_3$)1.40 (3H, t, $\underline{CH}_3CH_2O$), 4.40 (2H, q, $CH_3\underline{CH}_2$-O), 4.45 (2H, s, Ph-$\underline{CH}_2$-O-Ph), 5.10 (2H, s, Ph-$\underline{CH}_2$-CO-CO_2Et), 7.20 (2H, m, 5 and 6$\underline{H}^s$), 7.40 (5H, m, $\underline{Ph}$ -CH_2-O-Ph $\underline{H}^s$), 7.80 (1H, d $\underline{J}_{3,5}$ 2Hz - 3$\underline{H}$). $\underline{m/e}$ 344 (M + 1, 3%), 343 ($M^{+\cdot}$ 18), 295 (2), 270 (5), 243 (52), 200 (2), 165 (4), 120 (5), 105 (8), 91 (100 base), 77 (12), 65 (48). Metastables at 46.4 for 91 $\longrightarrow$ 65, corresponding to -26 and 34.1 for 243 $\longrightarrow$ 91 corresponding to $-CH_3C_6H_3NO_3$.

6-Benzyloxyindole-2-carboxylic acid (143)

A hot solution of hydrated ferrous sulphate (97.2 g) in water (190 cm^3) was added rapidly with stirring to a suspension of 4-benzyloxy-2-nitrophenylethylpyruvate (15.00 g) in a solution of aqueous ammonia made by adding concentrated aqueous ammonia solution (52 cm^3) to water (190 cm^3). The mixture was stirred mechanically and heated at 90 - 100° on a steam bath for 1 hour. During this time the ester dissolved and formed a dark brown solution. After 1 hour the mixture was heated at gentle reflux temperature for an additional 30 minutes.

The black mixture was filtered hot through sintered glass to remove the ferric oxide sludge. The filter cake was washed thoroughly with hot 2N ammonium hydroxide solution until the washings were colourless. The dark amber filtrate was chilled to 0 - 5° in an ice-bath and acidified by the dropwise addition of concentrated hydrochloric acid with continuous stirring. The acid was added at such a rate that the temperature did not exceed 20° and the crude product precipitated as a grey solid. This solid was filtered off, washed with a little cold water and dried in a vacuum oven at 60° for 4 hours to give (3.20 g).

Repeated extraction of the ferric oxide filter cake with hot dichloromethane, followed by drying over anhydrous sodium sulphate and evaporation of the combined organic layers under reduced pressure gave an orange solid. This was dissolved in 2N aqueous ammonia solution and acidified with concentrated hydrochloric acid at ice temperature as before, to yield a further crop, which after collection, washing and drying, weighed (0.63 g). The combined products were crystallized from benzene to give a white microcrystalline solid (3.74 g, 32%) m.p. 185 - 186° (lit;[89] m.p. 185 - 186°) both with decomposition λ_{max} (ε) 245 shoulder (20, 150), 248 (25,260), 312 (16,420)nm. ν_{max} 3310 (N-H st), 3,100-2,600 (O-H bonded and alkyl), 1700 (C=O st), 1160 and 1070 (C-O-C and CH_2-O-Ar st), 920 (O-H def), 860 and 800 (1,2,4-aromatic substitution pattern for indole A ring), 770 and 720 (aromatic substitution pattern for the monosubstituted benzyl ring).

$(CD_3)_2SO$ 5.05 (2H, s, Ph-C$\underline{H}_2$-O), 7.0 (1H, s, -3$\underline{H}$), 7.40 (1H, s broad - exchanges with D_2O-N$\underline{H}$), 7.45 (7H, m, 4 and 5H^s and the benzyl aromatic $\underline{H}^s$), 7.60 (1H, s broad - exchanges with D_2O-COO$\underline{H}$), 8.00 (1H, d $\underline{J}_{7,3}$ 2Hz - 7$\underline{H}$). $\underline{m/e}$ 268 (M + 1 0.2%), 267 ($M^{+\cdot}$ 1), 243 (3), 239 (3), 223 (1), 213 (62),197 (5), 139 (2), 126(2), 110 (16), 91 (100 base), 77 (4), 66 (11). Metastables at 186.25 for 267-223 corresponding to -44-CO_2 and 47.86 for 91-66.

Attempted decarboxylation of 6-benzyloxyindole-2-carboxylic acid (Method A).

The acid (1.0 g) was ground to an intimate mixture with copper chromite (0.1 g - obtained commercially) and the mixture suspended in quinoline (15 cm^3). The suspension was heated to 215° and held

at this temperature for 2 hours with stirring in a thermostatic oil bath.

The mixture was cooled to room temperature and poured into water (50 cm^3). The dark solution was repeatedly extracted with ether (5 x 30 cm^3). The combined ether extracts were washed with 2N-hydrochloric acid (2 x 50 cm^3), saturated sodium hydrogen carbonate solution (2 x 50 cm^3), followed by water (3 x 25 cm^3). The ether extract was then dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a dark brown coloured solid. Both T.L.C. and mass spectral analysis showed this to consist of starting material and product. Chromatography on a column of neutral alumina eluting with diethylether/petroleum ether (b.p. 40 - 60°) in the ratio of 8:2 separated the two components. The product was separated from the elute as a cream solid, which was crystallized from hot petroleum ether (b.p. 40 - 60°) as small white plates (0.207 g, 25%). m.p. 111 - 112° (lit;[89] m.p. 111 - 112°. λ_{max} (ε) 245 (10,850), 293 (6,820), 305 shoulder (6,660) and 315 shoulder (5,580) nm. ν_{max} 3,400 (N-H st), 1630 (N-H def), 1600 and 1590 (d-Ar-H st), 1230 and 1100 ($Ph-O-CH_2$ anti symmetric st and symmetric st), 870 and 815 (1,2,4-aromatic substitution on the indole ring), 760 and 715 (monosubstitution on the phenyl ring) cm^{-1}. δ ($CDCl_3$) 5.02 (2H, s, Ph-$\underline{CH}_2$-O), 6.70 (1H, d $\underline{J}_{3,2}$ 4Hz - $\underline{3H}$), 7.30 (1H, d x 2 $\underline{J}_{5,4}$ 8Hz and $\underline{J}_{5,7}$ 2Hz - $\underline{5H}$), 7.40 (7H, m, 4 and $7H^s$ and the benzyl aromatic H^s), 7.65 (1H, d $\underline{J}_{2,3}$ 4Hz - $\underline{2H}$), 8.0 (1H, s broad, exchanges with D_2O - $\underline{NH}$). m/e 224 (M + 1, 3%), 223 ($M^{+\cdot}$ 22), 132 (87), 116 (9), 104 (11), 91 (100 base), 87 (28), 78 (12).

Cold Ehrlich's reagent (p-dimethylaminobenzaldehyde) gave a red colouration which changed to green on heating as noted by Burton and Stoves.

6-Benzyloxyindole (144, Optimised Method B).

The acid (2.0 g) was mixed with glycerol (20 cm^3) and heated at 215 - 230° in an oil bath with stirring until gas evolution ceased (about 25 minutes). The mixture was cooled to room temperature and poured into water (100 cm^3). The product was extracted with ether (3 x 100 cm^3). The combined ether extracts were dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a dark red, oily, liquid. Extraction with hot petroleum ether (b.p. 80 - 100°) (6 x 50 cm^3) gave a pale yellow solution that on concentration to low bulk, and cooling to 0° for several hours gave a white solid (0.559 g, 33.5%) m.p. 110 - 111° (lit.[89] m.p. 111 - 112°). Vacuum sublimation at a pressure of 0.3 mm furnished the product as a white amorphous solid. Crystallization then provided small white plates from petroleum ether (b.p. 40 - 60°) (0.431 g, 26%) m.p. 111 - 112°.

On repeating this experiment the temperature was reduced to 140 - 160° and the time of heating to 10 minutes. This gave a cleaner product that could be crystallized directly from petroleum ether and consequently a better yield of (0.782 g, 47%)from 2 g of acid (143).

The aqueous phase from either of the above reactions, when concentrated to low bulk and acidified with concentrated hydrochloric acid gave a precipitate of the unchanged acid. In the case where 2 g of acid was used (0.78 g, 39%) was returned.

4-Methyl-3-nitroaniline

A solution of sodium polysulphide was prepared by dissolving crystalline sodium sulphide ($Na_2S.9H_2O$) (40 g) in water (150 cm^3) and adding finely powdered sulphur (10 g). The mixture was warmed until a clear solution was obtained.

A mixture of 2,4-dinitrotoluene (25 g) and water (200 cm^3) was heated to gentle reflux under nitrogen with continuous mechanical stirring.

The sodium polysulphide solution was added dropwise to the refluxing and stirring solution over a period of 30 - 45 minutes. When addition was complete, stirring and refluxing were continued for a further 20 minutes. The mixture was allowed to cool to room temperature and the solid filtered off, washing with cold water. The product was transferred to a mixture of concentrated hydrochloric acid (35 cm^3) and water (150 cm^3) and refluxed for 15 minutes. The desired aniline dissolved, leaving sulphur and a small amount of unchanged starting material. The solution was filtered and the filtrate cooled to 10 - 15^o by the addition of crushed ice. Concentrated ammonia solution was then

added until the mixture was basic, while maintaining the temperature below 15° by means of an ice bath. The amine precipitated as a brown amorphous solid which was filtered off, drained dry, and recrystallised from 95% aqueous ethanol to give 4-methyl-3-nitroaniline as shining amber plates (13.20 g, 63.2%) m.p. 78 - 79° (lit;[225] 78 - 79°). λ_{max} (ε) 205 (6,400), 237 (14,000), 375 (960) nm. ν_{max} 3,450 and 3,375 (d NH_2 st), 1618 (br Ar-H and NH_2 def), 1510 ($ArNO_2$ St), 880 and 810 (1,2,4-aromatic substitution pattern) cm^{-1}. δ ($CDCl_3$) 2.48 (3H, s, $C\underline{H}_3$), 3.84(2H, sbr, $N\underline{H}_2$) 6.80 (1H, d x 2 $\underline{J}_{6,5}$ 8Hz and $\underline{J}_{6,2}$ 2Hz - 6$\underline{H}$), 7.10 (1H, d $\underline{J}_{5,6}$ 8Hz - 5$\underline{H}$), 7.25 (1H, d $\underline{J}_{2,6}$ 2Hz - 2$\underline{H}$). m/e 152 ($M^{+\cdot}$, 100% base), 135 (88), 107 (79), 77 (83). Metastable at 119.9 for 152 - 135. This experiment was repeated several times.

4-Methyl-3-nitrophenol (138, Method C).

To water (75 cm^3) was cautiously added concentrated sulphuric acid (101 g, 55 cm^3) with continuous mechanical stirring, the hot solution was cooled to 40° and finely powdered 4-methyl-3-nitroaniline (35 g, 0.209 mol) added. Crushed ice (120 g) was then added, and the mixture stirred until all the aniline had been converted into its sulphate and a homogeneous paste resulted. The paste was cooled to 0 - 5° in an ice/salt freezing mixture and a cold solution of sodium nitrite (18 g, 0.260 mol) in water (40 cm^3) was added over a period of 30 minutes maintaining the temperature between 0 - 7° with continuous stirring. When addition was complete, stirring was continued for a further 15 minutes and the solution tested with starch-iodide paper to

give an immediate and permanent colouration. Urea (25 g) was then added to destroy the excess nitrous acid. When gas evolution ceased the solution was filtered into a chilled receiver.

While the diazotisation was in progress concentrated sulphuric acid (165 cm^3) was cautiously added to water (150 cm^3) with continuous mechanical stirring. The sulphuric acid solution was heated just to boiling and the diazonium solution added dropwise to the refluxing, stirring solution over a period of 30 minutes. When addition was complete the heating was continued for a further 5 minutes. The flask was then cooled somewhat, and placed in ice-water. Stirring was continued until cold in order to obtain a homogeneous crystal-magma. The crude crystals were filtered off, dissolved in hot dilute hydrochloric acid (1:1 v/v concentrated acid/water), decanted from the residual dark oil, filtered while hot and chilled to 0^o in an ice-bath. Light amber crystals separated, these were collected, dried in air overnight and recrystallized from 95% aqueous ethanol to give amber plates (22.3 g, 63%) m.p. 79 - 81^o (lit;[92] 78 - 79^o). All the other spectral data were identical with those obtained for this compound from (Method A. p. 241).

This experiment was repeated several times in order to build up a small stock of 4-methyl-3-nitrophenol and implement the rest of the synthesis.

4-Methoxy-2-nitrotoluene (148).

To a solution of 4-methyl-3-nitrotoluene (25 g, 0.156 mole) in 2N-sodium hydroxide solution (150 cm^3) was added dimethylsulphate (30 g, 22.5 cm^3, 0.238 mol). The dark red solution was heated under reflux for three hours, cooled to room temperature and concentrated aqueous ammonia solution (S.G 0.88, 150 cm^3) added, to destroy the excess dimethylsulphate. The solution was allowed to cool to room temperature with constant mechanical stirring. The stirred solution was then rendered acidic by the cautious, dropwise addition of concentrated hydrochloric acid, at less than 20^o in an ice-bath. The product was then extracted with dichloromethane (4 x 200 cm^3), the combined organic layers were dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a dark red oil. The crude product was distilled at 108^o/0.4 mm pressure to furnish the pure ether as a pale yellow oil (23.0 g, 84.3%). A small sample that was frozen by cooling to 0^o and then allowed to warm up to room temperature, melted at 17^o (lit;[226] 17^o), λ_{max} (ε) 239 (5,440), 268 (3,200), 339 (1,600) nm ν_{max} 3,00 and 2,900 (Ar-H st), 2975 (Ar-CH_3 st sharp), 1510 (Ar-NO_2 st), 1030 (Ar-OCH_3 st), 810 and 880 (1,2,4-aromatic substitution pattern) cm^{-1}. δ ($CDCl_3$) 2.50 (3H, s, $C\underline{H}_3$), 3.80 (3H, s, $OC\underline{H}_3$), 7.05 (1H, d x 2, $\underline{J}_{5,6}$ 7Hz, $\underline{J}_{5,3}$ 2Hz - 5$\underline{H}$), 7.15 (1H, d $\underline{J}_{5,6}$ 7Hz - 6$\underline{H}$), 7.50 (1H, d $\underline{J}_{3,5}$ 2Hz - 3$\underline{H}$). m/e 167 ($M^{+\cdot}$, 58%), 150 (91), 134 (11), 122 (100, base), 106 (25), 91 (47), 78 (46), and metastable ions at 134.7 for 167 ⟶ 150 and 99.2 for 150 ⟶ 122.

Methylation of 4-methyl-3-nitrophenol (Method B)

Anhydrous ether (30 cm^3) was placed in a three necked, round bottom flask (250 cm^3 capacity), fitted with a dropping funnel (50 cm^3 capacity), nitrogen inlet, motor driven stirrer (with seal) and a reflux condenser, protected with a calcium chloride guard tube.

Sodium (freshly cut into small pieces and degreased with petroleum ether (b.p. 40 - 60°) under a stream of dry nitrogen), 0.75 g = 0.0326 g atom) was added quickly against a positive nitrogen pressure.

A slow stream of dry nitrogen was passed through the flask, above the surface of the stirred liquid, and a mixture of methanol (30 cm^3) and anhydrous ether (20 cm^3) was added dropwise at such a rate as to maintain a gentle reflux. When all the sodium had dissolved (approximately one hour), the nitrogen was shut off and the solution allowed to cool to room temperature.

The ether was removed by evaporation under reduced pressure, and 4-methyl-3-nitrophenol (5.0 g, 0.0326 mol), dissolved in methanol (20 cm^3) was added to the solution of sodium methoxide, dropwise over a period of 30 minutes. The temperature being maintained below 30° by means of an ice-bath. Methyl iodide (4.63 g, 2.03 cm^3 or 1 mol equivalent) in methanol (15 cm^3) was then added, and the mixture heated at reflux temperature for 4 hours, cooled to 40° and the excess methanol removed by evaporation under reduced pressure.

The residue was distilled at 108° under a pressure of 0.3 mm to give pure 4-methoxy-2-nitrotoluene as a pale yellow liquid (0.53 g, 9.8%).

Increasing the reaction period to 8 hours and/or using excess methyl iodide failed to improve the yield of product.

Preparation of Chromyl chloride[227]

A solution of chromium trioxide (15.0 g, 0.010 mol) in water (15 cm^3) was placed in a three necked flask of (250 cm^3 capacity), fitted with a dropping funnel, mechanical stirrer and a combined gas outlet/thermometer attachment. The mixture was cooled to 0° in an ice-salt bath and concentrated hydrochloric acid (33 cm^3) added slowly, maintaining the temperature below 10°. When addition was complete, concentrated sulphuric acid (45 cm^3) was added, dropwise with constant stirring, at such a rate that the temperature remained at less than 10°. When addition was complete stirring was continued for a further five minutes and then the mixture was transferred to a separating funnel. The lower layer of chromyl chloride was withdrawn and distilled through an 18 inch Vigreux column at 116° under normal pressure to give (14.63 g, 63%) of pure product as a dark red liquid.

The chromyl chloride was immediately placed in a glass stoppered bottle which was stored in the refrigerator.

Etards oxidation of 4-methoxy-2-nitrotoluene and the isolation of 4-methoxy-2-nitrobenzoic acid (149).

4-Methoxy-2-nitrotoluene (5.0 g, 0.029 mol), was dissolved in dichloromethane (20 cm^3) and cooled to -20° in a dry-ice/methanol bath. An ice-cold solution of chromyl chloride (4.64 g, 0.03 mol) in the same solvent (30 cm^3) was then added portionwise to the stirred solution over a period of one hour. After addition was complete the solution was stirred for a further 30 minutes at -20°, and a saturated solution of sodium hydrogen sulphite (75 cm^3) was added. The two phase system was stirred vigorously for 10 minutes and then allowed to separate. The dichloromethane layer was collected, dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a cream coloured solid. T.L.C. and mass spectral analysis of this product showed it to consist of a mixture of the desired 4-methoxy-2-nitrobenzoic acid, with the acid as the major component.

In order to separate these components the mixture was dissolved in dichloromethane (30 cm^3) and shaken with a saturated aqueous solution of sodium carbonate (30 cm^3) for 15 minutes. The aqueous phase was then separated and made just acidic by the dropwise addition of 2N-hydrochloric acid, maintaining the temperature at less than 20° by means of an ice-bath. 4-Methoxy-2-nitrobenzoic acid precipitated as a cream coloured amorphous solid. Crystallization from the minimum amount of hot methanol gave stout white needles (3.65 g, 62%), m.p. 196-197° (literature[228] m.p. 195-196°). λ_{max} (ε) 210 (11,500), 228 (12,050), 255 (7,000), 340 (850) nm. ν_{max} 3,300 -

2,500 (H-bonded O-H st), 1690 (C=O st), 1600 (Ar C-H st), 1545 (NO_2 st), 1310 (C-O st and O-H def (coupled), 1260, (C-O-OCH_3 st), 1090, 1040, 940 (O-H def, out of plane), 900, 860, 810, 800 (Ar, C-N st and 1,2,4-aromatic substitution pattern) cm^{-1}. δ ($CDCl_3$) 3.85 (3H, s, $OC\underline{H}_3$), 7.05 (1H, d x d $\underline{J}_{5,6}$ 7Hz $\underline{J}_{5,3}$ 2Hz - 5$\underline{H}$), 7.20 (1H, d $\underline{J}_{3,5}$ 2Hz - 3$\underline{H}$), 7.90 (1H, d $\underline{J}_{6,5}$ 7Hz - 6$\underline{H}$), 10.1 (1H, s-broad, exchanges with D_2O-$CO_2\underline{H}$). m/e 137 ($M^{+\cdot}$, 100% base), 122 (11), 107 (13), 94 (15), 77 (14). Metastable at 83.60 for 137 $\longrightarrow$ 107 corresponding to -OCH_3.

The dichloromethane phase from the previous separation was washed with saturated brine (2 x 50 cm^3), followed by water (2 x 75 cm^3) and dried over anhydrous sodium sulphate. The dry extract was evaporated under reduced pressure to give 4-methoxy-2-nitrobenzaldehyde as a white amorphous solid. Crystallization from 95% aqueous ethanol gave short white needles (0.59 g, 10.1%) m.p. 95 - 96° (literature[109] 95.5 - 96°).

All the other spectral data were identical with those quoted for this compound prepared by the successful route detailed on (p. 259).

As this result clearly shows that over oxidation occurs in this reaction we reduced the time taken to 30 minutes, but similar results were obtained. Addition of the sodium hydrogen sulphite solution after only five minutes reaction, and rapid separation of the products also failed to improve the yield. Finally the reaction

was repeated as before, but for only two minutes followed by addition of aqueous sodium hydrogen sulphite solution and rapid steam distillation of the aldehyde from the reaction mixture, but this gave similar unsatisfactory results.

4-Methoxy-2-nitrobenzaldehyde (Method A).

A. 4-Methoxy-2-nitrobenzaldiacetate (150)

A solution of 4-methoxy-2-nitrotoluene (20.0 g = 0.12 mol) in glacial acetic acid (240 g = 228 cm^3) and acetic anhydride (244.8 g = 226 cm^3 = 2.4 mol) was cooled to 0^o in an ice-salt bath. Concentrated sulphuric acid (34 cm^3 = 0.6 mol) was added dropwise to the mechanically stirred solution over a period of twenty minutes, so that the temperature did not rise above 5^o. The mixture was then allowed to cool to 0^o and chromic trioxide (40 g = 0.4 mol) was added portionwise over a period of one hour, at such a rate that the temperature did not exceed* 5^o. When addition was complete stirring was continued for a further thirty minutes at 0 - 5^o. The dark green solution was then poured onto crushed ice (1.6 Kg) with stirring. Ice-water was added to give a total volume of (2.5 l) and the mixture set aside for two hours.

The heavy white precipitate of product was filtered off, washing with cold water until the filtrate appeared colourless. The solid was collected and resuspended in cold 2% sodium carbonate solution (200 cm^3) with vigorous stirring for five minutes. The diacetate was again filtered off and washed with cold water (2 x 30 cm^3) followed by cold ethanol (20 cm^3). The near pure product was crystallized from ethanol/water 3:2 (60 cm^3) to give the pure diacetate as a white microcrystalline solid (19.20 g,

58.2%), m.p. 110 - 111°. λ_{max} (ε) 222 (15,200), 272 (2,880), 330 (1,440) nm. ν_{max} 1765 and 1740 (O-CO-CH_3 st), 1520 (NO_2 st), 1080 and 1020 (Ar-OCH_3 st) cm^{-1}. δ ($CDCl_3$) 2.1 (6H, s, CH_3COO), 3.9 (3H, s, OCH_3), 7.2 (1H, d x 2 $J_{5,6}$ 8Hz $J_{5,3}$ 2Hz - 5H), 7.55 (1H d $J_{6,5}$ 8Hz - 6H), 7.68 (1H, d $J_{3,5}$ 2Hz - 3H), 8.10 (1H, s, $HC(OCOCH_3)_2$). m/e 283 ($M^{+\cdot}$ 2%), 255 (11), 224 (4), 181 (33), 165 (25), 151 (100, base), 134 (16), 106 (23), 75 (21). * If the temperature exceeds 5° at this point, a poor yield results.

B. 4-Methoxy-2-nitrobenzaldehyde (146).

A mixture of 4-methoxy-2-nitrobenzaldiacetate (16.0 g = 0.0565 mol), water (36 cm^3), ethanol (36 cm^3) and concentrated sulphuric acid (6 cm^3) was heated at reflux temperature for thirty minutes. The hot solution was rapidly filtered through a pre-heated sintered glass funnel, and the filtrate chilled in an ice-bath. White needles separated, and were collected, washed with cold water (2 x 25 cm^3), drained and dried in a vacuum oven at 40° for five hours. This first crop weighed (9.2 g). The filtrate was diluted with water (120 cm^3), whereupon a second crop was obtained, which after drying as before, weighed (0.6 g). The product was almost pure but it was recrystallized from 95% aqueous ethanol to give (9.7 g, 94.8%) of white micro needles m.p. 95 - 96°, (lit[109] 95.5 - 96°) λ_{max} (ε) 219 (8,920), 248 (10,000), 277 (5,800), 337 (1,760) nm. ν_{max} 1690 (ArHC=O st), 1600 (Ar-H st), 1510 (Ar-NO_2 st), 1070 and 1030 (d, Ar-OCH_3 def), 880 and 810 (1,2,4-aromatic substitution pattern), 780 (OC-H def) cm^{-1}. δ ($CDCl_3$) 4.0 (3H, s, OCH_3), 7.10 (1H, d x 2 $J_{5,6}$ 7Hz $J_{5,3}$ 2Hz - 5H) 7.50 (1H, d, $J_{3,5}$ 2Hz - 3H), 7.95 (1H, d $J_{6,5}$ 7Hz - 6H), 10.10 (1H, s, CHO). m/e 181 ($M^{+\cdot}$, 16%), 150 (100, base),

135 (25), 108 (23), 95 (18). Metastable at 125.97 for 181 - 150 and 99.20 corresponding to 181 - 135, for $-OCH_3$ and $-NO_2$ respectively.

These experiments were repeated several times, in order to build up a small stock of 4-methoxy-2-nitrobenzaldehyde.

Attempted preparations of 4-methoxy-2-, β -dinitrostyrene[110] (147)

4-Methoxy-2-nitrobenzaldehyde (7.24 g, 0.040 mol) was dissolved in methanol (25 cm^3) with mechanical stirring. Nitromethane (2.44 g, 2.16 cm^3 = 0.04 mol) was added and the mixture cooled to 0^o in an ice/salt-bath. Sodium hydroxide (1.7 g, 0.0425 mol) was dissolved in water (2cm^3), this solution was allowed to cool to room temperature and then diluted to (5 cm^3) with ice-water. The sodium hydroxide solution was then added to the stirred nitromethane mixture, dropwise, at such a rate that the temperature was maintained between 10 - 15^o.*

During the addition a white precipitate formed and the mixture thickened to such an extent that it became difficult to stir , and methanol (10 cm^3) was added to convert the pasty mass to a more easily stirred slurry. When addition was complete the mixture was allowed to stand for fifteen minutes and then it was converted to a clear solution by the addition of ice-water (300 - 350cm^3), containing crushed ice.

This pale yellow solution was then poured in a thin stream into a previously prepared mixture of concentrated hydrochloric acid (10 cm^3) and water (15 cm^3) with constant stirring. A pale

yellow solid separated immediately and was filtered off on sintered glass, washing with water (3 x 50 cm^3) portions. The solid was drained dry and dissolved in dichloromethane. The organic phase was washed with saturated sodium hydrogen carbonate solution (2 x 50 cm^3), saturated brine (2 x 50 cm^3) and water (3 x 75 cm^3). The dichloromethane phase was then dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a pale yellow solid (7.1 g).

T.L.C. and mass spectral analysis showed this solid to be unchanged 4-methoxy-2-nitrobenzaldehyde. All the other spectral date were also identical with those quoted on (p. 260) for the preparation of this compound.

Repeating this reaction using various concentrations of reagents and reaction times failed to give any of the desired product.

Finally the base used was changed to sodium methoxide but this also failed to produce the desired product, thus we changed our method of synthesis to the technique successfully employed by Woodward[109], and detailed on (p. 263).

* The first few drops of sodium hydroxide solution should be added cautiously to the nitromethane mixture since, after a short induction period there is a considerable evolution of heat. The rest of the sodium hydroxide solution may then be added more rapidly.

The reaction is rapid above 10^{o} and the optimum temperature range for the successful reaction with other substrates is said to be 10 - 15^{o} (Organic Synthesis, collected volumes 1 1941, 641).

4-Methoxy-2, β -dinitrostyrene (147)

A mixture of 4-methoxy-2-nitrobenzaldehyde (20.0 g = 0.110 mol), nitromethane (20 cm^3 = 17.74 g = 0.290 mol), ammonium acetate (8.0 g) and glacial acetic acid (80 cm^3) was stirred and heated to 60^{o} under a protective nitrogen atmosphere. The mixture was held at this temperature with constant stirring for fifteen minutes and the hot, dark orange solution, slowly poured onto crushed ice (400 g), with vigorous stirring. The product was precipitated as an oily orange solid. The ice was allowed to melt at room temperature and the solid stirred for two hours before filtering.

The filter cake was drained and dried in a vacuum oven for two hours at 60^{o} to give an amorphous orange powder (21.6 g). This powder was dissolved in chloroform, washed with saturated sodium bisulphite solution (aqueous sodium metabisulphite 54 g/l) (2 x 50 cm^3), twice with saturated brine followed by drying over anhydrous sodium sulphate. The dried solution was heated at reflux temperature with decolourising carbon for ten minutes, filtered and evaporated under reduced pressure, at less than 35^{o} (darkening was seen to occur at higher temperatures in chloroform), to small volume. Petroleum ether (b.p. 40 - 60^{o}) was added to bring about crystallization. This material was collected and recrystallized from methanol to give the pure product as yellow

needles (16.42 g, 66.3%) m.p. 103 - 104° (lit[109] 103 - 104°)

λ_{max} (ε) 223 (12,800), 273 (7,360), 323 (7,680), 345 (7,200) nm.

ν_{max} 3,100 ($R^1CH=CHR^2$ st), 1630 (Ar-C=C st), 1540 and 1500 ($C=CHNO_2$ and $ArNO_2$ st), 1280, 1060 and 1030 (Ar-OCH_3 st), 970 ($R^1CH=CHR^2$C-H def out of plane trans)cm^{-1}. δ($CDCl_3$) 3.98 (3H, s, $OC\underline{H}_3$), 7.25 (1H d x 2 $\underline{J}_{5,6}$ 8Hz $\underline{J}_{5,3}$ 2Hz - 5H), 7.42 (1H, d, J 14Hz H C=$CHNO_2$), 7.60 (1H, d $\underline{J}_{6,5}$ 8Hz-6H), 7.62 (1H, d $\underline{J}_{3,5}$ 2Hz - 3H), 8.45 (1H, d J 14Hz CH=C$\underline{H}NO_2$). The coupling constant indicates a trans stereochemistry of the double bond, as expected. m/e 224 ($M^{+\cdot}$, 48%), 150 (60), 133 (45), 122 (100 base), 107 (38), 95 (50), 78 (62), Metastables at 99.22 for 150 → 122 and 73.97 for 122 → 95.

6-Methoxyindole (Method A).

4-Methoxy-2,β-dinitrostyrene (15.0 g) was dissolved in ethylacetate (170 cm^3). To this solution, ethanol (20 cm^3), glacial acetic acid (25 cm^3) and 10% palladium-charcoal (1.5 g) were added. The mixture was hydrogenated at a hydrogen pressure of 3.5 Kg/cm^2 for twenty five minutes at room temperature.

When the reaction was complete the catalyst was filtered off and the filtrate added with stirring to a mixture of ether (150 cm^3) and saturated sodium hydrogen carbonate solution (120 cm^3). The two-phase system was stirred virorously for 1½ hours and then allowed to settle. The layers were separated and the ether phase was extracted several times with water (4 x 60 cm^3), to remove ethanol, acetic acid and ammonium acetate.

The ether solution was then dried over anhydrous sodium carbonate and concentrated under reduced pressure at less than 30° to approximately (25 cm^3). Petroleum ether (b.p. 40 - 60°) was then added to the cloud point, and the crude product precipitated as a brown crystalline mass. This material was collected and dissolved in dry ether. The ether solution was passed through a short column of basic alumina (75 g). The eluted ether was concentrated to very low volume under reduced pressure, once again at less than 30° and petroleum ether (b.p. 40 - 60°) added. The cloudy solution was stored in ice at 0° for 1½ hours whereupon pure white plates of 6-methoxyindole precipitated. The product was collected and dried in a vacuum desiccator to give (8.36 g, 85%) m.p. 91 - 92° (lit [109] 91 - 92°) λ_{max} (ε) 220 (29,100), 267 (4,100), 292 (5,370) n.m. ν_{max} 3,390 (N-H st), 1635 (C=C st), 1600 (Ar-H st), 1260 (Ar-O-CH_3 antisymmetric st), 1165 and 1030 (Ar-OCH_3 st) cm^{-1}. δ ($CDCl_3$) 3.80 (3H, s, $OC\underline{H}_3$), 6.45 (1H, d $\underline{J}_{3,2}$ 4Hz - 3$\underline{H}$), 6.75 (1H, d x 2 $\underline{J}_{5,4}$ 8Hz and $\underline{J}_{5,7}$ 2Hz - 5$\underline{H}$), 6.82 (1H, d $\underline{J}_{7,5}$ 2Hz - 7$\underline{H}$), 7.10 (1H, d $\underline{J}_{2,3}$ 4Hz - 2$\underline{H}$), 7.5 (1H, d $\underline{J}_{4,5}$ 8Hz - 4$\underline{H}$), 7.95 (1H, s broad, exchanges with D_2O - N$\underline{H}$). $\underline{m/e}$ 147 ($M^{+\cdot}$, 100% base), 132 (91), 116 (7), 104 (30), 87 (6), 78 (11). Metastables at 118.53 for 147 → 132 corresponding to -15, -CH_3 and 81.93 for 147 → 104. (Found: C, 75.4; H, 6.1; N, 9.5. C_9H_9NO requires C, 75.53; H, 6.34; N, 9.51.

6-Methoxyindole (Method B).

A mixture of pure crystalline 4-methoxy-2,β-dinitrostyrene (2.1 g = 0.0093 mol), iron powder (7.0 g), ethanol (14 cm^3) and glacial acetic acid (14 cm^3) was stirred mechanically and warmed on a water-bath until the reaction became vigorous, it was then removed from the heat and allowed to moderate. Stirring

and heating were then continued, the reaction mixture being kept just below the boiling point (90 - 100°) by gentle heating and cooling if necessary. After ten minutes the mixture thickened and the colour changed from brown to greenish-grey, heating was continued for a further five minutes and the mixture then poured into a suspension of sodium metabisulphite (7.0 g) in water (150 cm^3) with stirring.

The green suspension was extracted with ether (5 x 50 cm^3), the combined organic layers were washed with 50% sodium hydrogen carbonate solution (4 x 100 cm^3), followed by water (2 x 100 cm^3), dried over anhydrous sodium sulphate and stirred with decolourising carbon for five minutes at 30°. The hot solution was filtered and the ether evaporated under reduced pressure, at less than 30° to give a pale yellow oil. The oil was dissolved in dry ether and passed through a short column of basic alumina. The eluted ether was concentrated to very low bulk, under reduced pressure, once again at less than 30°, whereupon a pale brown crystalline mass separated. This crude product was recrystallized from the minimum quantity of 1:1 benzene/petroleum ether (b.p. 40 - 60°) to give pure white plates of 6-methoxyindole (858 mg, 62.3%) m.p. 91 - 92° (lit[109]; 91 - 92°). All the other spectral and analytical data were identical with those obtained for the product of (Method A).

4-Methoxy-2-nitrobenzaldehyde (Method B)

A solution of formaldoxime (10%) was prepared in a 5 l flange flask equipped with a reflux condensor, mechanical stirrer and pressure equalizing dropping funnel. Paraformaldehyde (34.5 g) and

hydroxylamine hydrochloride,(79.0 g) were heated and stirred with water (510 cm^3) until a clear solution was obtained. Sodium acetate (153 g) was then added, and the reaction mixture was kept under gentle reflux with stirring for 15 - 20 minutes. The solution was cooled to 10^o and sodium sulphate (3 g) and cupric sulphate pentahydrate ($CuSO_4.5H_2O$) (18.75 g) were added followed by a solution of sodium acetate (495 g) in water (540 cm^3) with stirring.

To this solution of formaldoxime, was added an ice-cold solution of 4-methoxy-2-nitro benzene diazonium chloride which had been prepared in advance as follows.

4-Methoxy-2-nitroaniline (126 g) was suspended with stirring in a mixture of concentrated hydrochloric acid (171 cm^3), water (150 cm^3), and crushed ice (300 g). A solution of sodium nitrite (52.5 g) in water (75 cm^3) was added slowly with continual stirring keeping the temperature between 0 - 5^o by means of an ice-salt bath. When addition was complete stirring was continued for a further 10 minutes and the solution tested with starch-iodide paper to give an immediate and permanent colouration. Urea (30 g) was then added portion-wise to destroy excess nitrous acid and after gas evolution ceased the solution was filtered through sintered glass into a chilled receiver. A solution of sodium acetate (92.5 g) in water (155 cm^3) was added to the dark red diazonium solution until it was neutral to congo-paper.

This solution of 4-methoxy-2-nitrobenzene diazonium chloride was added dropwise, below the surface of the formaldoxime solution

with the aid of a long stemmed dropping funnel, with vigorous mechanical stirring. The oxime of 4-methoxy-2-nitrobenzaldehyde was precipitated as a dark brown tar. The nitrogen liberated during the reaction tended to cause heavy foaming. This was controlled by the addition of a small quantity ($\sim$ 20 - 30 cm^3) of ether. After addition was complete, stirring was continued for one hour, during which time the tar coalesced into one large lump. The mixture was then acidified to congo with concentrated hydrochloric acid and left to stand overnight.

The supernatant liquor was then decanted from the tarry oxime, to which ferric ammonium sulphate (900 g) and water (1.5 l) were added. This mixture was heated at reflux temperature for 40 minutes, and then distilled with steam. The first fraction of distillate contained a considerable amount of m-nitroanisol (orange-yellow solid m.p. 37 - 38^{o}). This was separated from the desired aldehyde by dissolving the material in chloroform and washing with saturated sodium bisulphite solution. The aqueous phase was made just basic with 10% sodium hydroxide solution and extracted with dichloromethane. The organic phases were combined and dried over anhydrous sodium sulphate. The dry solution was evaporated under reduced pressure to give the product as a pale yellow amorphous solid.

The steam distillation was continued until no more aldehyde separated from the cold distillate (approximately 16 hours). The aldehyde was then collected, washed with cold water, drained and dried at 40^{o} in a vacuum oven for 6 hours. The crude material was

combined and recrystallized from 95% aqueous ethanol to give white micro needles (81.84 g, 55.3%), m.p. 95 - 96° (lit[109] 95 - 96°). All the other physical data were identical with those quoted for the product of method A on (p. 265).

4-Methoxy-2-aminotoluene (153)

4-Methoxy-2-nitrotoluene (10.0 g) was dissolved in ethanol (200 cm^3). Adam's platinum oxide catalyst (0.2 g) was added and the mixture hydrogenated at room temperature for 3½ hours, under a hydrogen pressure of 3.5 Kg/cm^2.

The catalyst was filtered off and the ethanol evaporated under reduced pressure at less than 35°, to give an amber coloured amorphous solid. Crystallization from ethanol at -20° over a period of 16 hours afforded the pure product as shining amber cubes (7.88 g, 96%) m.p. 39 - 40° (lit[229] m.p. 38°) λ_{max} (ε) 247 (5,920), 290 (7,000) n.m. ν_{max} 3,400 and 3,340 (d - NH_2 st), 1610 (NH_2 def), 1600 (Ar-H st), 1280 (C-N st), 1030 ($ArOCH_3$ st), 860 and 800 (1,2,4-aromatic substitution pattern) cm^{-1}. δ ($CDCl_3$) 2.05 (3H, s, $C\underline{H}_3$), 3.55 (2H, s broad, exchanges with D_2O - $N\underline{H}_2$), 3.70 (3H, s, $OC\underline{H}_3$), 6.25 (1H, d, $\underline{J}_{3,5}$ 2Hz - 3$\underline{H}$), 6.30 (1H, d x 2 $\underline{J}_{5,6}$ 7Hz $\underline{J}_{5,3}$ 2Hz - 5$\underline{H}$), 6.90 (1H, d $\underline{J}_{6,5}$ 7Hz - 6$\underline{H}$). m/e 138 (M + 1, 8%), 137 ($M^{+\cdot}$, 100 base), 136 (M - 1, 57), 122 (7), 120 (6), 109 (9), 107 (13), 94 (12), 93 (9), 77 (10). Metastables at m/e 108.60 corresponding to 137 → 122 or -15, $-CH_3$ and 86.72 for 137 → 109 corresponding to -28 -CO.

(E) and (Z)-4-Methoxy-2-formamidotoluene (154).

4-Methoxy-2-aminotoluene (7.0 g = 0.05 mol) was mixed to a slurry with formic acid ((95 - 97%) 8.0 cm^3 = 9.76 g = 4.0 molar equivalents). The mixture was heated on a steam bath at 95° under a short condenser for 4 hours, during which time the solids dissolved to form a deep amber coloured solution. This solution was cooled to room temperature and allowed to stand overnight, the excess formic acid was then distilled off at 75° under a pressure of 0.2 mm to give a buff coloured solid.

The solid was dissolved in dichloromethane (100 cm^3) and washed with saturated sodium bicarbonate solution (4 x 100 cm^3), followed by saturated brine (2 x 100 cm^3). The dichloromethane phase was dried over anhydrous sodium sulphate and heated with decolourising carbon at reflux temperature for 5 minutes. The carbon was filtered off, and the dichloromethane evaporated under reduced pressure to give an off-white solid. Crystallization from the minimum quantity of methanol gave a white microcrystalline solid (7.06 g, 84%) m.p. 83 - 84° λ_{max} (ε) 214 (13,900), 240 (6,800), 285 (3,000) n.m. ν_{max} 3,300 (broad N-H st), 1650 (broad NHC$\underline{O}$ st), 1600 (Ar C H st), 1100 and 1040 ($ArOCH_3$ st), 860 and 810 (1,2,4-aromatic substitution pattern) cm^{-1}. The p.m.r. spectrum shows two closely spaced sets of signals, indicating the ($\underline{E}$) and ($\underline{Z}$) forms.

δ ($CDCl_3$) 2.18 and 2.00 (2 x 3H, s, C$\underline{H}_3$), 3.75 and 3.78 (2 x 3H, s, OC$\underline{H}_3$), 6.6 (2 x 1$\underline{H}$, 2 (d x 2) $\underline{J}_{5,6}$ 8Hz $\underline{J}_{5,3}$ 2Hz - 5$\underline{H}^s$), 7.05 (2 x 1$\underline{H}$, 2 x d $\underline{J}_{6,5}$ 7Hz - 6$\underline{H}^s$), 7.56 (2 x 1H, 2 x d $\underline{J}_{3,5}$ 2Hz - 3$\underline{H}^s$), 8.10 (2 x 1$\underline{H}$, s broad, exchanges with D_2O N$\underline{H}^s$), 8.40 and 8.35 (2 x 1$\underline{H}$, s,

$NCO\underline{H}^{s}$). m/e 166 (M + 1 8%), 165 ($M^{+\cdot}$, 81), 148 (13), 137 (81), 136 (100, base), 122 (16), 106 (24), 94 (20), 93 (20), 77 (22). Metastable at 112.09 for (165 ⟶ 136) corresponding to -28 or -CO.

Cyclization of (E) and (Z)-4-methoxy-2-formamidotoluene

Commercial t-butanol (55 cm^3) was placed in a three necked, round bottomed flask (250 cm^3 capacity), fitted with a nitrogen inlet, motor driven stirrer (with seal) and a reflux condenser.

Potassium (freshly cut into small pieces and degreased with petroleum ether (b.p. 40 - 60°) under a stream of dry nitrogen) (2.55 g = 0.065 g atom or 1.48 molar equivalents) was added portion-wise to the stirred alcohol against a small positive nitrogen pressure. The addition was carried out at such a rate that the reaction did not become vigorous. A slow stream of dry nitrogen was passed through the flask, above the surface of the stirred mixture and the flask was warmed on a steam bath until all the potassium had dissolved.

(E) and (Z)-4-Methoxy-2-formamidotoluene (6.0 g, 0.044 mol) was added and brought into solution by stirring the hot mixture. The stirrer was removed and the excess t-butanol distilled off under nitrogen. The residue was heated on a sand bath at 350 - 360° for 20 minutes under nitrogen and then allowed to cool in a stream of nitrogen. The cool residue was decomposed by the addition of water (50 cm^3), and the mixture steam distilled.

The distillate was extracted with ether (4 x 50 cm^3) portions. The ether extracts were combined and shaken with cold 2M-hydrochloric acid (2 x 50 cm^3), water (50 cm^3) and 5% sodium carbonate solution (50 cm^3). The ether solution was then dried over anhydrous sodium sulphate and stirred with decolourising carbon (0.2 g) at room temperature for 5 minutes. The carbon was filtered off, and the ether distilled under reduced pressure to give a brown solid. This solid was chromatographed on a short column of basic alumina, eluting with dry ether. The first band to emerge from the column was collected and the ether once more removed by evaporation under reduced pressure to give a tan coloured solid. Crystallization from petroleum ether (b.p. 40 - 60^o) gave a white microcrystalline solid (532 mg, 10%) m.p. 91 - 92^o (lit[109] m.p. 91 - 92^o). The other physical data were identical with those quoted for this compound on (p. 265).

Reducing the severity of the reaction conditions to 180 - 200^o for 30 minutes did nothing to improve the quality of the initial product or the yield.

The Preparation of 4 and 6-methoxy-2-amino-α-chloroacetophenones (157) and (158).

The following reactants were mixed together under reduced pressure with the aid of a vacuum line and anhydrous conditions.

Boron trichloride (2.93 g = 2.2 cm^3 = 0.025 mol) was added to dry benzene (10 cm^3) with stirring. To this was added a solution of 3-methoxyaniline (3.08 g = 2.81 cm^3 = 0.025 mol) in dry benzene (30 cm^3),

dropwise with stirring at 0^{o} in an ice-bath. Chloroacetonitrile (2.26 g = 1.90 cm^3 = 0.030 mol) was then added, followed by titanium tetrachloride (3.05 cm^3 = 0.0275 mol). The resulting mixture was heated at reflux temperature for 3 hours under dry nitrogen. After cooling to room temperature the mixture was rendered acidic by the addition of ice-cold 2N-hydrochloric acid (25 cm^3). The acidic solution was then warmed at 80^{o} for 30 minutes and cooled to room temperature. The pH of the cooled mixture was adjusted to 3 - 4 with 2N-sodium hydroxide and the solution extracted with dichloromethane (5 x 40 cm^3). The combined organic layers were washed with water (3 x 30 cm^3), dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a pale yellow oil. The oil was dissolved in benzene and passed through a short column of basic alumina, to remove a polar fraction. The benzene elute was evaporated under reduced pressure to give a pale straw coloured oily liquid. (2.30 g, 45%).

A sample of the product oil was dissolved in chloroform to form a 5% solution and analysed by G.L.C. With a column temperature of 180^{o} and nitrogen as the carrier gas at a pressure of 1.4 Kg/cm^2 the retention times for the 4 and 6-methoxy isomers were 2.5 and 3.5 minutes respectively. The peak areas suggest a product ratio of 54:46, in favour of the desired 4-methoxy isomer.

This was confirmed by analysis of the relative intensities of the corresponding methylene protons in the 100 MHz ^1H n.m.r. spectrum which is reproduced in (Figure 3).

The two sets of methylene protons, corresponding to the 4 and 6-methoxy isomers are clearly defined. The 4 — methoxy isomer is in slightly greater abundance and occurs at lower field than its 6-methoxy counterpart.

The methylene group of the 6-methoxy isomer experiences less deshielding due to the electron releasing influence of the adjacent methoxylgroup. The position of the aromatic protons can be rationalized in the same way.

Attempted Separation of 4 and 6-methoxy-2-amino-∝chloroacetophenones (157)and (158).

T.L.C. on basic alumina using benzene/petroleum ether (b.p. 40 - 60°)in the ratio of 7:3 showed only a slight difference in Rf values. Column chromatography was carried out using the same optimised system as for the T.L.C. analysis but it was not possible to achieve a clean separation of the two components. The first band to emerge from the column was collected and the solvent evaporated under reduced pressure to give a white solid. T.L.C. and G.L.C. once again suggested a mixture, and ^{1}H n.m.r. confirmed that the product contained both the 4 and 6-methoxy isomers. The material was enriched in the desired 4-isomer but not sufficiently for subsequent attempts at fractional crystallization from benzene to be effective.

Attempts to separate these isomers by the use of basic alumina preparative layer plates, eluting with a mixture of benzene and petroleum ether (b.p. 40-60°) in a ratio of 3:2 also failed to give a clean separation. This result is at variance with that of

Sugasawa et al.,[113] but these authors failed to specify which type of chromatographic support they employed in their separation. Further attempts with a number of different combinations of solvent systems and supports also failed, so we decided to cyclize the mixture without separation as follows.

Preparation of 4 and 6-methoxy indoles from the corresponding 2-amino-∝-chloroacetophenones

To a mixture of 4 and 6-methoxy-2-amino-∝-chloroacetophenones (0.5 g = 0.0025 mol) in dioxane (5 cm^3) containing water (0.5 cm^3) was added sodium borohydride (50 mg = 0.0013 mol) and the solution heated at reflux temperature for 4 hours. The mixture was cooled to room temperature and the solvent evaporated under reduced pressure, water (25 cm^3) was added and the mixture extracted with dichloromethane (4 x 50 cm^3). The combined dichloromethane extracts were dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a pale yellow coloured oil. Crystallization from diethyl ether/petroleum ether (b.p. 40 - 60°) in the ratio of 8:2 at low temperature gave a white microcrystalline solid (312 mg, 85%). T.L.C. and ^1H n.m.r. showed it to be the expected mixture of 6 and 4-methoxy indoles in the ratio of 2:1 respectively. The ^1H n.m.r. spectrum of this mixture exhibited the following signals for the methoxyl groups. δ ($CDCl_3$) 3.80 (3H, s, -6-methoxyindole OCH_3), and 3.90 (3H, s, -4-methoxyindole OCH_3). Integration of the spectrum, showed the peak area of the resonance due to the 6-methoxyindole to be twice that of the 4-isomer. This enrichment of the 6-methoxy component may indicate that some fractional crystallization takes place.

Attempted separation of 4 and 6-methoxyindoles

T.L.C. using a variety of systems, indicated very similar Rf values. The two best were diethylether/petroleum ether (b.p. 40 - 60°) in the ratio of 7:3 and benzene/diethylether in the ratio of 6:4, both on basic alumina. With both of these systems a very slight separation was observed on T.L.C. plates. However, column chromatography with these systems failed, in both cases, as did preparative basic alumina plates. An enriched mixture was obtained in all cases.

We next prepared the N-acetyl derivatives of 4 and 6-methoxy-2-amino-α-chloroacetophenones.

Preparation of 4 and 6-methoxy-2-acetamido-α-chloroacetophenones (159) and (160)

The mixed 4 and 6-isomers (1.5 g) were dissolved in dichloromethane (15 cm^3) and acetic anhydride (15 cm^3) added. The solution was warmed at 80° for 30 minutes with stirring and then cooled to room temperature. The bulk of the acetic anhydride was removed by evaporation under reduced pressure and the residue partitioned between 10% aqueous sodium hydrogen carbonate solution (50 cm^3) and dichloromethane (3 x 50 cm^3). The combined organic layers were dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a pale amber oil. The oil was chromatographed on a short column of silica gel, eluting with benzene/ethylacetate in the ratio of 6:4, to remove a very polar component. The first band to emerge from the column was collected and the solvents evaporated under reduced pressure to give a cream coloured amorphous solid (1.50 g, 83%).

Separation of 4 and 6-methoxy-2-acetamido-∝-chloroacetophenones

The mixed N-acetyl derivatives prepared in the previous experiment were separated on silica gel preparative plates, eluting with a mixture of benzene and ethyl acetate in a ratio of 6:4 at a loading of (100 mg) per plate. The desired isomer ran as the first band, and after 18 hours development it was recovered and extracted with hot dichloromethane. The solvent was evaporated under reduced pressure to give a white solid that was crystallized from dichloromethane/ethanol in the ratio of 1:1 to give a white microcrystalline solid.; Five plates gave (244 mg or 79% recovery of the 4-isomer) m.p. 130 - 131° (lit [113] 130 - 131°). ν_{max} 3251 (N-H st), 1700 (NH $\underline{CO}CH_3$ st) and 1657 ($\underline{CO}CH_2Cl$ st) cm^{-1}. δ ($CDCl_3$) 2.25 (3H, s, $\underline{CH}_3CONH$), 3.90 (3H, s, $OC\underline{H}_3$), 4.68 (2H, s, $COC\underline{H}_2Cl$), 6.65 (1H, d x 2 $\underline{J}_{5,6}$ 8Hz $\underline{J}_{5,3}$ 2Hz - 5$\underline{H}$), 7.75 (1H, d $\underline{J}_{6,5}$ 8Hz - 6$\underline{H}$), 8.45 (1H, d $\underline{J}_{3,5}$ - 2Hz - 3$\underline{H}$), 11.65 (1H, s-broad, exchanges with D_2O - N$\underline{H}$).

Hydrolysis of 4-methoxy-2-acetamido-∝-chloroacetophenone

The N-acetyl derivative prepared in the previous experiment (200 mg), was dissolved in dry methanol (25 cm^3) which had previously been saturated with dry hydrogen chloride gas. The solution was heated at reflux temperature for 2 hours and then cooled to room temperature. The solvent was evaporated under reduced pressure to give a pale yellow oil.

Ice was added and concentrated aqueous ammonia solution (S.G. 0.880) was added dropwise with stirring until the mixture was basic. The product was extracted with dichloromethane (3 x 50 cm^3). The combined organic extracts were dried over anhydrous sodium sulphate and

evaporated under reduced pressure to give a white solid which was crystallized from iso-propanol/dichloromethane in the ratio of 1:1 to give pure 4-methoxy-2-amino-α-chloroacetophenone (157) (152 mg) 92%) m.p. 108 - 109° (lit[113] m.p. 107 - 108°) ν_{max} 3510 and 3375 (d - NH_2 st) 1650 (C=O st), 870 and 815 (1,2 4-aromatic substitution pattern) 740 (C-Cl st) cm^{-1}. δ ($CDCl_3$) 3·88 (3H, s, $O\underline{CH}_3$), 4·58 (2H, s, $\underline{CH}_2$), 6·10 (1H, d $\underline{J}_{3,5}$ 2Hz - 3$\underline{H}$), 6.25 (1H, d x 2 $\underline{J}_{5,6}$ 8Hz $\underline{J}_{5,3}$ 2Hz - 5$\underline{H}$), 6.45 (2H, s-broad, exchanges with D_2O - $N\underline{H}_2$), 7.52 (1H, d $\underline{J}_{6,5}$ 8Hz - 6$\underline{H}$).

Conversion of 4-methoxy-2-amino-α-chloroacetophenone (157) to 6-methoxyindole

This was achieved using (120 mg) of the amine prepared in the previous experiment and sodium borohydride (25 mg). The reaction conditions and purification procedures were exactly as quoted for the mixed isomers on (p. 275). This experiment gave pure 6-methoxyindole as white plates (74 mg, 84%) m.p. 91 - 92[9] (lit[109] m.p. 91 - 92°) u.v. I.R., ^1H n.m.r. and mass spectral data were all identical with those quoted on (p. 265).

3-Acetyl pyridine

Finely chopped and degreased sodium (44.4 g) was dissolved in excess absolute ethanol under anhydrous conditions. When the reaction was complete the ethanol was evaporated under reduced pressure to give sodium ethoxide (130g= 1.41 mol) as a white solid. A solution of methyl nicotinate (151.25 g = 1.25 mol) in ethyl acetate (208.5 g = 2.36 mol) was added with stirring. The amber solution was stirred for

one hour and then heated at reflux for ten hours after which it was allowed to cool overnight. The pale-yellow mass was dissolved in water (1.2 l) and the unreacted ethers removed by extraction with diethyl ether (6 x 100 cm^3). Dissolved ether was removed from the aqueous solution by warming at 50° on a water bath for twenty minutes, after which the solution was cooled and made acidic by the addition of concentrated hydrochloric acid (500 cm^3). Evolution of carbon dioxide began and the hydrolysis was completed by heating vigorously at reflux for 2½ hours. The solution was then cooled, made alkaline by the portionwise addition of potassium carbonate and extracted with diethyl ether (6 x 150 cm^3). The ether extracts were dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a pale yellow oil. The pure ketone was obtained by distillation at 112°/0.5 mm pressure, giving a colourless oily liquid (94.5g, 74.3%). λ_{max} (ε) 230 (13,250), 268 (3,780), 275 (3,540)nm. ν_{max} 1700 (C=O st), 1600 (Ar-H st), 1280, 1030, 960, 810, 760 cm^{-1}. δ ($CDCl_3$) 2.4 (3H, s, $C\underline{H}_3$), 7.2 (1H, q, 5-$\underline{H}$), 7.7 (1H, d, -4$\underline{H}$), 8.15 (1H, d, 6-$\underline{H}$), 8.4 (1H, s, -2$\underline{H}$). m/e $M^{+\cdot}$ 121 (100% base).

3-(1-Hydroxyethyl)pyridine (34).

To 3-acetylpyridine (25.0 cm^3) dissolved in ethanol (100 cm^3), sodium borohydride was added in small portions over a period of thirty minutes. The temperature was controlled to less than 30° by means of an ice-bath, and the reaction was judged to be complete by the observance of a constant ultraviolet spectrum. Distilled water (10 cm^3) was then added, and the ethanol removed by evaporation under reduced pressure The resulting yellow-green slurry was diluted with distilled water (75 cm^3), and extracted with dichloromethane in (5 x 20 cm^3) portions, the organic extracts were combined and dried

over anhydrous sodium sulphate. Removal of the solvent under reduced pressure gave a pale yellow oil, that was purified by distillation at 90° under a pressure of 0.3 mm to give the pure product as a colourless oily liquid (21.2 g, 95%). λ_{max} (ε) 263 (6,500) with shoulders at 256 (5,800) and 268 (4,700) n.m. ν_{max} 3,500 - 3,100 (H-bonded OH st), 2950 and 2,900 (CH_3-asymmetric st), 1610 and 1600 (d - Ar - H st - pyridine nucleus), 1410 (O-H def) cm^{-1}. δ ($CDCl_3$) 1.5 (3H, d, $C\underline{H}_3$), 4.9 (1H, q, C$\underline{H}$), 7.2 (1H, d x d $\underline{J}$ 7Hz and $\underline{J}$ 7Hz - 5$\underline{H}$), 7.7 (1H, d x d $\underline{J}_{4,5}$ 7Hz and $\underline{J}_{4,2}$ 2Hz - 4$\underline{H}$), 8.25 (1H, d x d $\underline{J}_{6,5}$ 7Hz and $\underline{J}_{6,2}$ 2Hz - 6$\underline{H}$), 8.40 (1H, d $\underline{J}_{2,4}$ 2Hz - 2$\underline{H}$), and 8.55 (1H, s-broad-exchanges with D_2O - O$\underline{H}$). m/e 123 ($M^{+\cdot}$ 60%), 108 (100, base), 80 (90), and a metastable ion at 59.26 for (108 ⟶ 80) or - CO.

3-(1-chloroethyl)pyridine (162)

To 3-(1-hydroxyethyl)pyridine (20 g) in dry benzene (50 cm^3) was added thionyl chloride (120 cm^3) dropwise, maintaining the temperature at 5 - 10° by means of an ice-bath. The mixture was evaporated under reduced pressure to give a sticky white jelly of the pyridinium hydrochloride. This was taken up in ice-water (50 cm^3) and washed with ether in (3 x 20 cm^3) portions. The aqueous solution was then made basic by the portion wise addition of solid sodium hydrogen carbonate with constant stirring, the rate of addition being adjusted so that the temperature could be maintained at less than 20° by means of an ice-bath. When just basic to litmus the aqueous solution was extracted with diethylether in (3 x 100 cm^3) portions. The ether solution was washed several times with water, dried over anhydrous sodium sulphate and evaporated under reduced pressure at a temperature not exceeding 30°, to yield

the chlorinated product as a mobile, unstable, light-yellow liquid, (22 g, 96%). ν_{max} 650 (C-Cl st) cm^{-1}. The mass spectrum was run at low temperature and gave the following data: m/e 141 ($M^{+\cdot}$, 34%), 126 (6), 106 (100, base), and 90 (11). The ^1H n.m.r. spectrum did not exhibit an OH signal. δ $[(CD_3)_2SO]$ 1.8 (3H, d, CH_3CH), 5.5 (1H, q, CH_3CH), 7.3 (1H, d x d J 7Hz and J 7Hz - 5H), 7.9 (1H, d x d $J_{4,5}$ 7Hz and $J_{4,2}$ 2Hz - 4H), 8.45 (1H, d x d $J_{6,5}$ 7Hz and $J_{6,2}$ 2Hz - 6H), 8.70 (1H, d $J_{2,4}$ 2Hz - 2H).

A D.M.S.O. spectrum of the starting material 3-(1-hydroxy ethyl)pyridine showed an OH doublet at 5.4 p.p.m.

3-[1-(3-Pyridyl)ethyl]-6-methoxyindole (161) Method A.

Magnesium turnings (0.86 g, 0.034 mol) were covered with dry ether (25 cm^3) under a protective nitrogen atmosphere. Anhydrous conditions were maintained throughout, and bromoethane (2.5 cm^3 ≈ 3.68 g = 0.034 mol) in dry ether (40 cm^3) was added with stirring (15 cm^3) all at once, to initiate the reaction, followed by the remainder at such a rate as to maintain a gentle reflux. When the reaction was complete, approximately 45 minutes, all the magnesium had dissolved leaving a grey solution of ethylmagnesium bromide.

Next the solution was cooled to 0 - 5° in an ice-salt bath and 6-methoxyindole (5.0 g, 0.034 mol) in dry ether (30 cm^3), was added dropwise over a period of 20 minutes, with stirring under nitrogen. To this mixture maintained at 0 - 5° was added 3-(1-chloroethyl)pyridine (4.81 g, 0.034 mol) in dry ether (10 cm^3), dropwise over a period of 10 minutes. Stirring was maintained at 5° for 4 hours, followed by

one hour at room temperature after which the mixture was set aside and left to stand at room temperature for a further 24 hours.

The solvents were then removed by evaporation under reduced pressure to give a sticky, orange coloured gum. The gum was cooled to 0 - 5° in an ice/salt-bath and dissolved in 2M hydrochloric acid (50 cm^3) with vigorous stirring. Ether (60 cm^3) was added and the layers separated. The aqueous layer was washed with ether (5 x 50 cm^3), and then adjusted to pH 8 - 9 with concentrated aqueous ammonia solution, added dropwise at less than 20° in an ice-bath. The basic solution was extracted with ether (5 x 40 cm^3), followed by dichloromethane in (4 x 40 cm^3) portions. The dichloromethane extracts were combined, dried over anhydrous sodium sulphate and heated to reflux for five minutes with decolourising carbon (200 mg). The hot solution was filtered and the dichloromethane evaporated under reduced pressure to give a yellow oil. The oil was triturated with dry ether to give an off-white solid. This product was crystallized from 95% aqueous ethanol to give a white microcrystalline solid (1.29 g, 15.1%) m.p. 166 - 167°, λ_{max} (ε) 221 (21,600), 251 (4,300), 260 (6,300), 272 (3,800), 281 (2,400) n.m. ν_{max} 3,180 (N-H st), 1620 (N-H def), 1600 (Ar and py C- H st), 1165 and 1020 (Ar-OCH_3 st) cm^{-1}. δ [$(CD_3)_2SO$] 1.62 (3H, d, $C\underline{H}_3CH$), 3.80 (3H, s, $OC\underline{H}_3$), 4.35 (1H, q, $CH_3C\underline{H}$), 6.7 (2H, m, 5 and 7 indole $\underline{H}^s$) 7.1 - 7.4 (3H, m, 2 and 4 indole $\underline{H}^s$ and pyridine 5$\underline{H}$), 7.6 (1H, d x 2, $\underline{J}$ 6Hz and $\underline{J}$ 2Hz - pyridine 4$\underline{H}$), 8.15 (1H, d x 2 $\underline{J}$ 6Hz and $\underline{J}$ 2Hz - pyridine 6$\underline{H}$), 8.55 (1H, d $\underline{J}$ 2Hz - 2$\underline{H}$) 9.8 (1H, s broad, exchanges with D_2O N$\underline{H}$). m/e 252 ($M^{+\cdot}$, 73%), 237 (100, base), 222 (12), 212 (15), 174 (10), 134 (13), 106 (67), 78 (15). Metastable at 222.89 for (252 ⟶ 237)

corresponding to loss of CH_3. (Found: C, 76.2; H, 6.4; N, 11.2; $C_{16}H_{16}N_2O$ requires: C, 76.2; H, 6.3; N, 11.1%).

Due to the low yield of this experiment we attempted to optimize the conditions in the following ways:

1. The experiment was repeated as before, except that the temperature was held at 0^{o} for the first hour, followed by stirring at room temperature for a further twenty four hours.

2. The temperature was maintained at 10 - 15^{o} for the first hour and then the experiment was worked up immediately.

The products of experiments 1 and 2 were however, the same as in the original procedure, thus we carried out experiment 3.

3. After stirring the reaction mixture at 5^{o} for one hour it was heated at its reflux temperature for two hours, but the dichloromethane extract from this experiment gave only 4% of the desired product and a large quantity of resinous material, that could not be separated by chromatography.

A considerable amount of unreacted 6-methoxyindole was recovered from the ether extracts of all these reactions.

1,1-Bis(3-indolyl)-1-(3-pyridyl)ethane (2)

Indole (23.4 g = 0.2 mol) and 3-acetylpyridine (12.1 g = 0.1 mol) were dissolved in glacial acetic acid (50 cm^3) and finely powdered zinc chloride (0.1 g) added. The mixture was heated at reflux temperature for 3½ hours, and the orange solution cooled to room

temperature. Water (50 cm^3) was added and the solution basified, by the dropwise addition of concentrated aqueous ammonia (S.G. 0.880), with constant stirring at less than 15° in an ice-bath. A cream coloured solid formed rapidly. When the solution was basic to litmus the product was filtered off and crystallized from ethanol to give a white microcrystalline solid (31.38 g, 93%) m.p. 251 - 252° (lit[6] m.p. 251 - 252°) λ_{max} (ε) 223 (11,500), 264 (12,400), shoulder at 272 (12,000) and a second shoulder at 280 (10,400), n.m. ν_{max} 3,400 (N-H st), 1610 (N-H def), 1600 (Ar and py C-H st), 1280, 1240, 1180, 1140, 1060, 1040, 840, 760 (1,2 aromatic substitution) cm^{-1}. δ [$(CD_3)_2SO$] 2.25 (3H, s, $\underline{CH}_3$), 6.80 (2H, d $\underline{J}$ 4Hz - indole $2\underline{H}^s$), 6.85 - 7.40 (9H, m, indole aromatic $\underline{H}^s$ and pyridine -5$\underline{H}$), 7.65 (1H, d x 2 $\underline{J}_{4,5}$ 7Hz $\underline{J}_{4,2}$ 2Hz - pyridine - 4$\underline{H}$), 8.35 (1H, d x 2 $\underline{J}_{6,5}$ 7Hz $\underline{J}_{6,4}$ 2Hz - pyridine - 6$\underline{H}$), 8.50 (1H, d $\underline{J}_{2,4}$ 2Hz - pyridine - 2$\underline{H}$), 10.9 (2H, d broad, exchanges with D_2O - N-$\underline{H}^s$). m/e 338 (M + 1, 9%), 337 ($M^{+\cdot}$, 37), 323 (25), 322 (100, base), 260 (2), 259 (9), 243 (5), 205 (4), 168.5 (s, ½ mass ion), 161 (4). Metastable at 307.6 for (337 ⟶ 322) corresponding to -15 or $-CH_3$.

1-(3-indolyl)-1-(3-pyridyl)ethylene (166)

1,1-Bis-(3-indolyl)-1,-(3-pyridyl)ethane (30.0g = 0.089 mol) was pre-dried in a vacuum oven at 120° for 4½ hours. The dried material was finely powdered and vacuum pyrolysed at 280 - 285° under a pressure of 0.3 mm, in a system fitted with a nitrogen bleed. Heating was supplied from a sand bath and continued for twenty minutes, during which time, the solid melted to a smooth dark liquid and indole distilled onto the condenser as a crystalline white solid. After twenty minutes no more indole distilled, so the dark mixture was allowed to cool to room temperature under nitrogen.

The solid mass was dissolved in 2N-hydrochloric acid (100 cm^3) and the acidic solution extracted with ether (6 x 50 cm^3), to remove traces of indole. The mixture was then carefully basified by the dropwise addition of concentrated aqueous ammonia to the stirring solution at 10 - 15° in an ice-bath. A somewhat tarry, buff-coloured precipitate separated and was collected, dissolved in dichloromethane followed by drying over anhydrous sodium sulphate. The dry dichloromethane solution was heated at reflux temperature for 10 minutes with decolourising carbon, filtered hot and evaporated under reduced pressure to give an off-white sticky solid.

Direct crystallization from ethanol, ethanol-water, and ethyl acetate, failed to purify this compound. The T.L.C. showed one major and three minor spots, which separated well on basic alumina using 7:3 ethylacetate/petroleum ether (b.p. 60 - 80°). The crude product was chromatographed on a short column of basic alumina using this solvent system, the desired compound running well ahead of the minor products. Evaporation of the elute containing the first band to emerge from the column gave a white amorphous solid, that could now be crystallized from ethanol to give 1-(3-indolyl)-1-(3-pyridyl) ethylene as a white microcrystalline solid (1.97 g, 10.1%) m.p. 145 - 146° λ_{max} (ε) 225 (25,400), 268 (11,600), 282 (7,330), 293 (3,840) n.m. ν_{max} 3,200 (N-H st), 1650 and 1640 (close doublet N-H def and $R^1R^2C{=}CH_2$ st), 1600 (Ar-H st), 910 ($R^1R^2C{=}CH_2$ def), 750 and 700 (aromatic substitution) cm^{-1}. δ [$(CD_3)_2SO$] 4.40 and 4.70 (2 x 1H, 2 x d $\underline{J}$ 2Hz $>C = C<^{H}_{H}$), 6.85 (1H, s, indole - 2$\underline{H}$), 6.90 - 7.05 (4H, m indole 4,5,6 and 7$\underline{H}^s$), 7.20 (1H, m, pyridine -5$\underline{H}$), 7.8 (1H, d x 2 $\underline{J}_{4,5}$ 7Hz $\underline{J}_{4,2}$ 2Hz pyridine 4$\underline{H}$), 8.35 (1H, d $\underline{J}_{6,5}$ 7Hz -

pyridine - 6H), 8.6 (1H, d $J_{2,4}$ 2Hz - pyridine - 2H), 9.6 (1H, s broad, exchanges with D_2O - NH). m/e 220 ($M^{+\cdot}$, 11%), 193 (100 base).

Due to the poor yield of this experiment we attempted to optimize the conditions in the following ways:

1. The temperature of the experiment was reduced to 250 - 260°, to minimise charring, but on working up in the above manner approximately 20% by weight of starting material was isolated and no significant increase in yield of desired product was obtained.

2. The time of pyrolysis was reduced to ten minutes, but once again the reaction was found to be some 50% incomplete.

3. The reaction was carried out using the original temperature of 280 - 285° but extending the pyrolysis time to 45 minutes. On working up this experiment our fears were confirmed. The only extractable material was a dark, resinous gum, which failed to yield to the chromatographic techniques applied successfully above.

1-(3-indolyl)-1-(3-pyridyl)ethane (113).

1-(3-indolyl)-1-(3-pyridyl)ethylene (1.80 g = 0.008 mol) was dissolved in a mixture of ethanol (40 cm^3) and glacial acetic acid (10 cm^3), (this material is not sufficiently soluble in ethanol alone) and hydrogenated over a 10% palladium-carbon catalyst (0.2 g) using a hydrogen pressure of 4.2 Kg/cm^2 for 4½ hours at room temperature.

The catalyst was filtered off, and the solvents evaporated under reduced pressure. The residue was partitioned between dichloromethane and dilute aqueous ammonia (1:1 water/concentrated SG. (0.88)). The organic phases were combined, dried over anhydrous magnesium sulphate and evaporated under reduced pressure to give a light yellow oil, which on trituration with ether gave an off-white solid. Crystallization from 95% aqueous ethanol furnished 1-(3-indolyl)-1-(3-pyridyl)ethane as a white microcrystalline solid (1.68 g, 93%). m.p. 173 - 174° (lit;[54] 173 - 174°) λ_{max} (ε) 225 (13,600), 272 (7,800), 287 (6,200), 296 (5,200) n.m. ν_{max} 3,140 (N-H st), 1610 (N-H def), 1600 (Ar-H st), 1260, 1160, 1115, 1040 and 1030 d, 810, 750 and 710 (aromatic substitution) cm^{-1}. δ [$(CD_3)_2SO$] 1.60 (3H, d $\underline{J}$ 4Hz - $C\underline{H}_3CH$), 4.30 (1H, q, $\underline{J}$ 4Hz - $CH_3C\underline{H}$), 6.82 (1H, s, indole - 2$\underline{H}$), 7.0 - 7.1 (1H, m, pyridine - 5$\underline{H}$), 7.20 - 7.4 (4H, m, indole 4,5,6 and 7H^s), 7.65 (1H, d x 2 $\underline{J}_{4,5}$ 7Hz $\underline{J}_{4,2}$ 2Hz - pyridine 4$\underline{H}$), 8.35 (1H, d $\underline{J}_{6,5}$ 7Hz - pyridine 6$\underline{H}$), 8.60 (1H, d $\underline{J}_{2,4}$ 2Hz - pyridine - 2$\underline{H}$), 10.90 (1H, s broad, exchanges with D_2O N$\underline{H}$) m/e 223 (M + 1, 10%), 222 ($M^{+\cdot}$, 68), 207 (100, base), 180 (7), 144 (13), 117 (10), 104 (11), 77 (7). Metastable at 193.01 for (222 ⟶ 207) corresponding to -15 or $-CH_3$.

Attempted condensation of indole and 3-acetylpyridine under basic conditions.

1. A round bottomed, three necked flask of (250 cm^3, capacity) equipped with a combined dropping funnel/gas inlet, mechanical stirrer (with seal) and a reflux condenser, protected by calcium chloride guard tubes was assembled hot, from oven dried apparatus and cooled in a stream of dry nitrogen. Dry dimethylsulphoxide

D.M.S.O. (40 cm^3) was placed in the flask and indole (2.0 g, 0.0171 mole) was added. 3-Acetylpyridine (2.06 g = 1.87 cm^3, 0.0171 mole) was dissolved in dry D.M.S.O. (10 cm^3) and added to the stirred solution. Sodium hydride ($\sim$ 2.0 g) was degreased in petroleum ether (b.p. 40 - 60°) under a stream of dry nitrogen. Dry sodium hydride (0.82 g, 0.0342 moles or 2 molar equivalents) was then added portionwise to the stirred mixture under a protective atmosphere of dry nitrogen. The rate of addition was adjusted so that the temperature did not exceed 25°. When addition was complete (approximately 15 minutes) the mixture was heated to 60° in a thermostatic oil bath, and held at this temperature with stirring for 3 hours, during which time the solution changed colour from orange to chocolate-brown.

The mixture was cooled to room temperature and the excess sodium hydride was destroyed by the cautious, dropwise addition of ethanol. When foaming ceased the ethanol was evaporated under reduced pressure and the residue poured into an excess of water (1½ litres). The solution was then adjusted to pH 7 with dilute hydrochloric acid. The neutral solution was extracted, first with diethylether in (4 x 150 cm^3) portions to remove any starting materials, followed by dichloromethane in (4 x 200 cm^3) portions. The dichloromethane extracts were combined, dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a small amount of a dark brown gum smelling strongly of D.M.S.O. and dimethylsulphide. This residue was heated on a rotatory evaporator at 60° under a pressure of 0.3 mm for 4 hours to remove the residual solvent. The solid

residue was examined by T.L.C. on silica plates eluting with a mixture of dichloromethane and petroleum ether (b.p. 40 - 60°) in a ratio of 3:2. This showed it to consist of a mixture of very polar polymeric products that would not give a satisfactory separation with this solvent system. Ethyl acetate/petroleum ether (b.p. 40 - 60°) in a ratio of 4:1 and chloroform/methanol 98:2 were also used with silica and neutral alumina plates, but without success.

It was felt that the failure to isolate any product from this reaction might be due to its solvation by dimethylsulphoxide in the aqueous phase. Therefore, experiment 2 was carried out.

2. The experiment was carried out as previously described in 1, except that the D.M.S.O. was removed by rotatory evaporation at 60° under a pressure of 0.3 mm for 4 hours before the work up procedure.

The residue was partitioned between water (500 cm^3) and ether in (4 x 150 cm^3) portions, followed by dichloromethane in (4 x 100 cm^3) portions. This, once again, gave a small quantity of brown gum. T.L.C. analysis of this residue using basic alumina or silica plates and eluting with a mixture of ethyl acetate and petroleum ether (40 - 60°) in a ratio of 4:1 again showed it to consist of a multicomponent mixture of high polarity compounds. Comparison with an authentic sample of 1,-(3-indolyl)-1-(3-pyridyl)ethylene, synthesized previously (see p. 284), showed that none of the desired compound was being formed. Evaporation of the ether extracts under reduced pressure gave a quantity of sticky solid. T.L.C. on neutral alumina plates, eluting with a mixture of ether and petroleum ether

(b.p. 60 - 80°) in a ratio of 3:2 proved it to contain a great deal of starting materials.

The results of experiment 2, lead us to speculate that the reaction conditions were not sufficiently energetic so experiments 3 and 4 were carried out.

3. The reaction was repeated as previously described in 2, except that the temperature was held at 60° for 16 hours. This reaction gave little starting material in the ether extracts but much polymeric gum in the dichloromethane fraction. However, T.L.C. analysis was carried out using the same type of plates and solvent system as for 2, but none of the desired product could be detected and the polymeric residue would not yield to attempts at chromatography.

4. The experiment was again repeated in the same manner as described previously for 2, except that this time the temperature was raised to 120°, and held at this temperature for only 1½ hours. Once again results were similar to those obtained in experiment 3.

2-Methylpropanilide

2-Toluidine (17.19 g, 17.1 cm^3, 0.160 mol) was dissolved in ethyl acetate (75 cm^3) and propionic anhydride (20.88 g, 20.6 cm^3, 0.160 mol), was added dropwise with stirring over a period of ten minutes, maintaining the temperature at less than 30° by means of an ice-bath. When addition was complete the reaction vessel was transferred to a thermostatic water bath and the mixture maintained at 40° for four hours under a short air condenser.

The solvent was then removed by evaporation under reduced pressure to give a sticky, cream coloured solid. This was dissolved in dichloromethane (100 cm^3) and the solution washed with saturated sodium hydrogen carbonate solution (2 x 50 cm^3) portions, followed by saturated brine (2 x 30 cm^3) and water (2 x 50 cm^3). The dichloromethane layer was then dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a white solid. This was crystallized from ethanol to give the pure product as white needles (25.1 g, 96%) m.p. 43 - 44° (lit.[230] m.p. 43 - 44°)

λ_{max} (ε) 211 (7,400) (shoulder), 236 (12,500), 262 (600) n.m.

ν_{max} 3,460 and 3,400 (d N-H st), 3,100 - 2,880 (alkyl C-H st), 1600 and 1630 (d C=O st), 1600 (Aryl C-H st), 1580 and 1560 (N-H in plane def), 1280, 1060, 760 (1,2 aromatic substitution) cm^{-1}

δ ($CDCl_3$) 1.8 (3H, t, $C\underline{H}_3CH_2$), 2.2 (3H, s, $C\underline{H}_3Ph$), 3.4 (2H, q, $C\underline{H}_2CH_3$), 6.8 - 7.8 (4H, m, aromatic $\underline{H}^s$), 8.10 (1H, s-broad, exchanges with D_2O-N$\underline{H}$). m/e 163 ($M^{+\cdot}$, 68), 106 (100, base), 91 (18), 77 (20).

2-Ethylindole

Commercial t-butanol (207 cm^3) was placed in a three necked, round bottom flask (1l capacity), fitted with a nitrogen inlet, motor driven stirrer (with seal) and a reflux condenser.

Potassium (freshly cut into small pieces and degreased with petroleum ether (b.p. 40 - 60°) under a stream of dry nitrogen) (9.50 g, 0.24 g atom or 1.75 molar equivalents) was added portion wise to the stirred alcohol against a small positive nitrogen pressure. The addition was carried out at such a rate that the reaction did not become vigorous. A slow stream of dry nitrogen was passed through

the flask, above the surface of the stirred mixture and the flask was warmed gently on a steam bath until all the potassium had dissolved.

2-Methylpropanilide (22.5 g, 0.138 mole) was added and brought into solution by stirring the hot mixture. The stirrer was removed and the excess t-butanol distilled off under nitrogen. The stirrer was replaced and the residue heated and stirred at 200 - 220° in an oil-bath (with thermostatic control) for 1½ hours under nitrogen and then allowed to cool in a stream of nitrogen.

The cool residue was decomposed by the addition of water (200 cm^3) and the mixture steam distilled. The distillate was extracted with diethylether in (4 x 150 cm^3) portions. The ether extracts were combined and shaken with cold 2M-hydrochloric acid (2 x 150 cm^3), water (2 x 150 cm^3) and 5% sodium carbonate solution (2 x 150 cm^3). The ether solution was then dried over anhydrous sodium sulphate and s tirred with decolourising carbon (0.2 g), at room temperature for five minutes. The carbon was filtered off, and the ether evaporated under reduced pressure to give a tan-coloured mass. This solid was crystallized (twice) from petroleum ether (b.p. 40 - 60°) to give shining white plates (13.69 g, 68.4%). This experiment was repeated using the same quantities and the products combined to give (28.2 g in total) m.p. 44 - 45° (lit.[111] m.p. 86 - 87°) λ_{max} (ε) 223 (27,200), 270 (inflextion) (7,100), 285 (5,400) n.m.

ν_{max} 3,120 (N-H st), 3,000 - 2,850 (alkyl C-H st), 1625 (N-H def), 1600 (Aryl C-H st), 1250, 1130, 1060, 900, 890, 760 cm^{-1}.

δ ($CDCl_3$) 0.9 (3H, t, $C\underline{H}_3CH_2$), 2.5 (2H, q, $CH_3C\underline{H}_2$), 6.3 (1H, s, -3$\underline{H}$),

7.0 - 7.5 (4H, m, aromatic $\underline{H}^{s}$), 10.1 (1H, s broad, exchanges with D_2O - N$\underline{H}$). m/e 145 ($M^{+\cdot}$ 80), 90 (100, base), 78 (26).

2-Ethylindol-3-yl-3-pyridyl ketone (174)

A. Nicotinyl Chloride*

Potassium nicotinate (25.0 g, 0.165 mole) was finely ground and dissolved in dry benzene (120 cm^3). The solution was cooled to 0 - 5° in an ice-bath and oxalyl chloride (7.58 g, 5.2 cm^3 = 0.1 mole or 0.6 molar equivalents) also in dry benzene (40 cm^3) was added dropwise to the stirred solution. Stirring was continued at 0 - 5° for 30 minutes and the solution was then brought to reflux temperature for a further 30 minutes. The solution was then cooled to (-5°) - (-10°) in an ice/salt freezing mixture and used directly for the following Grignard reaction.

* H.N. Wingfield[231] reports an 80% conversion to the acid chloride using this technique.

B. Grignard Reaction

Magnesium turnings (4.8 g, 0.198 mole or 1.5 molar equivalents - assuming an 80% conversion of the acid chloride) were covered with dry tetrahydrofuran (100 cm^3) under a protective nitrogen atmosphere. Anhydrous conditions were maintained throughout and bromoethane (21.56 g, 14.8 cm^3 or 0.198 moles) in dry T.H.F. (150 cm^3) was added with stirring, (50 cm^3) all at once, to initiate the reaction, followed by the remainder at such a rate as to maintain a gentle reflux. When the reaction was complete, approximately one hour, all the magnesium had dissolved leaving a clear, grey solution of ethylmagnesium bromide.

Next the solution was cooled to 0 - 5° in an ice-bath and 2-ethylindole (23.92 g, 0.165 mole) in dry T.H.F. (75 cm^3) was added dropwise over a period of 20 minutes with stirring under nitrogen, to form a clear amber-coloured solution.

This solution (maintained under nitrogen) was added dropwise with constant mechanical stirring to the solution of nicotinyl chloride prepared previously. The reaction was maintained at -10° by means of an ice-salt freezing mixture for the duration of the addition period (approximately 30 minutes). The mixture was then allowed to warm up to room temperature and stirring was continued overnight. The organometallic complex was hydrolysed by the careful addition of a saturated ammonium chloride solution (20 cm^3), and the T.H.F. removed by evaporation under reduced pressure. The resulting gum was extracted with chloroform in (4 x 100 cm^3) portions. The chloroform extracts were combined, dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a yellow-green amorphous solid. Crystallization from ethanol (twice), afforded the ketone as colourless prisms; (29.69 g, 72%). m.p. 199 - 201°.

λ_{max} (ε) 265 (15,400), 325 (11,900) n.m. ν_{max} 3125 (N-H st), 1625 (N-H def), 1600 (Aryl and pyridyl C-H st), 1595 (C=O st) cm^{-1}.

δ [$(CD_3)_2SO$] 1.15 (3H, t J_{7Hz} - $CH_2\underline{CH}_3$), 2.8 (2H, q, J 7Hz - $\underline{CH}CH_3$), 7.05 - 7.6 (5H, m, -indolyl 4,5,6 and 7$\underline{H}$ and pyridine 5$\underline{H}$), 8.0 (1H, d x 2 J 8Hz and J2Hz - pyridine 4$\underline{H}$), 8.7 (1H, d x t J 8Hz and J 2Hz - pyridine 6$\underline{H}$), 8.9 (1H, d J 2Hz - pyridine 2$\underline{H}$), 12.0 (1H, s, broad, exchanges with D_2O - N$\underline{H}$). m/e 250 ($M^{+\cdot}$, 9%), 225 (12), 210 (14), 172 (9), 163 (89), 130 (22), 107 (100, base), 90 (13), 78 (25).

(Found: C, 76.6; H, 5.5; N, 11.4. $C_{16}H_{14}N_2O$ requires C, 76.8; H, 5.6; N, 11.2%).

Photochemical Studies of 2-Ethylindol-3-yl-3-pyridyl ketone (174)

The ketone (174) (250 mg) was dissolved in methanol (1 l) and the solution placed in a photochemical reactor of the water cooled immersion well type. Stirring was provided by a flow of nitrogen from a glass capillary, and the ultraviolet light was generated by a high pressure mercury arc lamp. The progress of the reaction was monitored by ultraviolet spectroscopy.

The ultraviolet spectrum of the starting material exhibits:

λ_{max} (ε) 238 (15,400) 328 (11,900) n.m.

The ultraviolet spectrum of ellipticine is reproduced below for comparison.

λ_{max} (ε) 238 (22,000), 275 (32,600), 285 (74,900), 294 (72,200), and 330 (4,500) n.m.

Thus it can be seen that a change to the ellipticine skeleton would be obvious from the ultraviolet spectrum.

A sample of the solution was withdrawn every 2 hours for a period of 16 hours, but no change in the ultraviolet spectrum was observed.

Next the triplet sensitizer benzophenone (0.05 g) was included in the reaction mixture and irradiation continued. No change occurred in the ultraviolet spectrum for 18 hours, and then a shift to shorter wavelength was observed. After 20 hours the change was complete and

the ultraviolet spectrum now showed one band with an absorption maxima of λ_{max} 256 n.m., indicating that no ellipticine had formed, as you would expect a shift to longer wavelengths if this were the case; due to the extended conjugation in the aromatic, tetracyclic system.

T.L.C. analysis confirmed that no pyrido[4,3-b]carbazoles were present, but indicated seven components, four major and three minor. The mass spectrum of this mixture showed the four major components to have m/e values of: 223, 193, 137 and 123 respectively, indicating that the molecule had simply fragmented.

The experiment was again repeated, but using acetophenone as the triplet sensitizer, however, similar negative results were observed.

Next we prepared the N-acetyl derivative of (174) or 1-acetyl-2-ethylindol-3-yl-3-pyridyl ketone (176), in a bid to reduce the amidic character of the carbonyl group.

The experiment was repeated as before, but using (250 mg) of (176), however, this treatment produced no new products. T.L.C. analysis showed only starting material to be present after 20 hours irradiation.

A. Attempted acetylation of the ketone (174)

The ketone (174) (2.0 g) was dissolved in acetic anhydride (30 cm^3) with stirring, and the mixture heated at reflux temperature for 4 hours. The solution was cooled to 30° and the acetic anhydride evaporated under reduced pressure to give a brown gum. This was partitioned between water (100 cm^3) and chloroform in (3 x 50 cm^3) portions. The chloroform phases were combined and washed with saturated sodium hydrogen carbonate solution (2 x 50 cm^3), and saturated brine (2 x 50 cm^3). The chloroform extract was then dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a buff-coloured solid (1.97 g). T.L.C. analysis of this on silica plates, eluting with ethyl acetate/petroleum ether (b.p. 40 - 60°) in a ratio of 4:1 showed that the solid isolated was unchanged starting material.

B. 1-Acetyl-2-ethylindol-3-yl-3-pyridyl ketone (176)

The ketone (174) (2.0 g) was dissolved in a mixture of acetic anhydride (20 cm^3), ethyl acetate (15 cm^3) and triethylamine (2 cm^3), with stirring. The solution was heated at reflux temperature for a period of 8 hours,* cooled to 30° and the bulk of the solvents evaporated under reduced pressure. This gave a green-coloured gum smelling strongly of triethylamine. Thus, the residue was heated at 45° on a rotatory evaporator under a pressure of 0.5 mm for 3 hours. This removed the triethylamine, and the residue was partitioned between saturated sodium hydrogen carbonate solution (75 cm^3) and chloroform in (4 x 100 cm^3) portions†. The organic phases were combined, washed with saturated sodium chloride solution (2 x 100 cm^3) and dried over anhydrous sodium sulphate. The dry chloroform solution was evaporated under reduced pressure to yield a cream-coloured solid.

This was crystallized from the minimum amount of methanol to give colourless plates (2.03 g, 87%). m.p. 174 - 175°.

Notes: * T.L.C. analysis of the reaction mixture at periods of less than 7-8 hours showed that the reaction was incomplete. Using the same solvent system as quoted previously for method A the Rf values are: 0.82 for the starting material (174) and 0.34 for the product (176).

† If an emulsion forms at this point it can usually be broken by filtering the mixture through a pad of 'filter aid'.

λ_{max} (ε) 260 (15,000), 282 (13,200) and a shoulder at 290 (5,100), 325 (11,900) n.m. ν_{max} 1700 (CH_3 C=O N st), 1640 (C=O st), 1600 (Aryl and pyridyl C-H st) cm^{-1}. δ [$(CD_3)_2SO$] 1.25 (3H, t, J 7Hz - $CH_2\underline{CH}_3$), 2.80 (3H, s, $\underline{CH}_3CON$), 3.10 (2H, q, J 7Hz - $\underline{CH}_2CH_3$), 7.0 - 7.4 (4H, m, - indolyl 4,5,6 and 7H), 7.7 (1H, 2-superimposed doublets J 8Hz - pyridine 5H), 8.10 (1H, 2 x t J 7Hz and 2Hz - pyridine 4H), 8.7 (1H, 2 x t J 8Hz and J 2Hz - pyridine 6H), 9.0 (1H, d J 2Hz - pyridine 2H). m/e 292 ($M^{+\cdot}$, 30%), 267 (12), 252 (15), 249 (50), 225 (9), 210 (10), 207 (48), 163 (90), 107 (100, base), 90 (12), 78 (30).

(Found: C, 73.9; H, 5.5; N, 9.6. $C_{18}H_{16}N_2O_2$ requires C, 74.0; H, 5.5; N, 9.6%).

Attempted $N_{(A)}$-benzene sulphonation of 2-Ethylindol-3-yl-3-pyridyl ketone (174)

Method A

The ketone (174) (1.5 g, 0.006 mol) was dissolved in ethyl acetate (75 cm^3) containing benzene sulphonyl chloride (1.27 g, 0.0072 mole or 1.2 molar equivalents) and triethylamine (2 cm^3). The mixture was heated at reflux temperature for 4 hours and then cooled to 30^o. The bulk of the solvents were removed by evaporation under reduced pressure to give a yellow coloured gum. This gum was heated at 40^o on a rotatory evaporator under a pressure of 0.5 mm for 3 hours to remove the residual triethylamine. The residue was partitioned between water (100 cm^3) and dichloromethane in (4 x 50 cm^3) portions. The dichloromethane extracts were combined and washed with saturated sodium hydrogen carbonate solution (4 x 50 cm^3), and saturated brine in (2 x 50 cm^3) portions. The dichloromethane extract was then dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a cream-coloured solid. Crystallization from the minimum amount of ethanol gave (1.23 g) of white plates. However, T.L.C. analysis, infrared and mass spectra all showed the product to be simply the starting material (174).

Method B.

To dry dimethylsulphoxide (75 cm^3), maintained under an atmosphere of dry nitrogen was added sodium hydride (1.92 g, 0.08 mol - degreased with petroleum ether (b.p. 40 - 60^o), under a stream of dry nitrogen). The sodium hydride was added portionwise over a period of 5 minutes, to the stirred solution. When addition was complete stirring was continued for 10 minutes and the ketone (174) (2.0 g, 0.008 mol)

was added portionwise over a period of 10 minutes, with constant stirring under a nitrogen atmosphere, the temperature being maintained at less than 20^{o} by means of an ice/salt-bath. When addition was complete the solution was cooled to -10^{o} and a solution of benzenesulphonyl chloride (1.41 g, 1.0 cm^3, 0.008 mol) in dry dimethylsulphoxide (10 cm^3) was added dropwise at such a rate that the temperature remained below 0^{o}. When all the benzenesulphonyl chloride solution had been added the mixture was stirred at -5^{o} for 2 hours, and at room temperature for a further hour.

The solution was then cooled to 0^{o} in an ice/salt-bath and ethanol (40 cm^3) added with care, to destroy any residual sodium hydride. The solvents were then removed, first under reduced pressure and then at 0.3 mm. The resulting sticky solid was partitioned between dichloromethane (100 cm^3) and water (75 cm^3). The dichloromethane layer was separated and washed with saturated sodium hydrogen carbonate solution (2 x 40 cm^3), saturated brine (3 x 20 cm^3) followed by water (2 x 50 cm^3). The dichloromethane layer was then dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a light brown coloured solid. Crystallization from ethanol gave a white microcrystalline solid (1.94 g) m.p. 199 - 201^{o}.

T.L.C. and mass spectral analysis of this solid proved it to be unchanged starting material (174).

Preparation of Triethyloxonium tetrafluoroborate[232]

A three necked flask of (250 cm^3 capacity) equipped with a mechanical stirrer, dropping funnel and reflux condenser (provided with calcium chloride guard tubes) was assembled hot, from apparatus dried in an oven at 110°. The apparatus was cooled in a stream of dry nitrogen and sodium dried ether (50 cm^3), followed by freshly distilled boron trifluoride etherate (28.4 g, 25 cm^3 or 0.2 mol) were placed in the cool flask. Freshly distilled epichlorohydrin (14.0 g, 12 cm^3 or 0.151 mol), was then added dropwise to the stirred solution at a rate sufficient to maintain vigorous boiling (about 30 minutes were required for the addition). The mixture was refluxed for a further hour and then allowed to stand overnight under an atmosphere of nitrogen.

The product crystallized, and the supernatant ether was decanted under nitrogen. The crystalline product was washed with dry ether in (3 x 50 cm^3) portions and the flask transferred to a dry box, where the crystals were collected on a sintered glass funnel. The product was placed in a tared bottle, in a dry box, under nitrogen, to give (25.65 g, 91%) of colourless crystals, m.p. 91 - 92 with decomposition (lit.[232] m.p. 91 - 92 with decomposition.)

Attempted O-ethylation of 2-Ethylindol-3-yl-3-pyridyl ketone (174)

The ketone (2.0 g, 0.008 mol) was dissolved in dichloromethane (40 cm^3), with stirring under a protective atmosphere of oxygen free nitrogen. Triethyloxonium tetrafluoroborate (3.8 g, 0.02 mole or 2.5 molar equivalents) were added quickly from a nitrogen purged flask.

The mixture was stirred at room temperature for 1½ hours, during which time a heavy yellow-coloured precipitate settled out. This precipitate was filtered off, washing with dichloromethane (15 cm^3), followed by dry ether in (4 x 50 cm^3) portions. The solid was crystallized from the minimum amount of methanol to give (2.8 g, 96%) of pale-yellow coloured micro-crystalline solid. m.p 268 - 270°.

This product proved to be the salt 1-ethyl-3-(2-ethyl-3-indolyl-formyl)pyridinium borotetrafluoride (179).

The salt was water soluble and exhibited the following ^1H n.m.r. spectra.

δ [$(CD_3)_2SO$] 1.40 (3H, t, $C\underline{H}_3CH_2$ - indole), 2.85 (3H, t, $C\underline{H}_3CH_2$ - $N^{\oplus}$), 3.01 (2H, q, $CH_3C\underline{H}_2$ - indole), 4.50 (2H, q, $CH_3C\underline{H}_2$ - $N^{\oplus}$), 7.2 - 7.48 (3H, m, 5, 6 and 7 - indole $\underline{H}^S$), 7.52 (1H, d x 2 $\underline{J}$ 7Hz, $\underline{J}$ 2Hz - indole 4$\underline{H}$), 8.0 (2H, m, pyridine 4 and 5 $\underline{H}^S$), 9.20 (1H, d x 2 $\underline{J}$ 7Hz, $\underline{J}$ 2Hz - pyridine 6$\underline{H}$), 9.38 (1H, $\underline{J}$ 2Hz - pyridine 2$\underline{H}$), 12.1 (1H, s, broad, exchanges with D_2O - N$\underline{H}$).

Phenylglycine-o-carboxylic acid

A slurry of anthranilic acid (50 g, 0.36 mol) in water (34 cm^3) was prepared. Separately a solution of sodium hydroxide (15.4 g) in water (28.5 cm^3) was prepared and cooled to room temperature. The anthranilic acid slurry was then added to the sodium hydroxide solution with stirring until a clear solution was obtained. Chloroacetic acid (34.7 g, 0.36 mol) was dissolved in water (50 cm^3) and anhydrous sodium carbonate added to the stirred mixture with care, over a period of 10 minutes or until the solution was slightly basic to litmus. The mixture was maintained below 20° by means of an ice-bath, and constant stirring was maintained,

excessive foaming being controlled by the addition of a trace of ether. The resulting solution of the sodium salt of chloroacetic acid was added to the previously prepared solution of sodium anthranilate and the mixture heated to 40° and held at this temperature for 4 hours in a thermostatic water bath.

The mixture was allowed to cool to room temperature, whereupon it formed a solid mass. This was dissolved by the addition of a solution of sodium hydroxide (15 g) in water (400 cm^3). This solution was then carefully acidified by the dropwise addition of concentrated hydrochloric acid, stirring continually and maintaining the temperature below 30° by means of an ice-bath. The product precipitated as a light tan coloured solid, and was collected, washed thoroughly with cold water and dried in a vacuum oven at 50° for 5 hours, to give (59.5 g, 84%) of almost pure product m.p. 219° (lit.[233] 218 - 219°)

N-Acetylphenylglycine-o-carboxylic acid .

Phenylglycine-o-carboxylic acid (19.0 g, 0.097 mol) was added portionwise with stirring to a solution of sodium carbonate (16.6 g) in water (166 cm^3). When all the solid had dissolved, acetic anhydride (14.6 cm^3, 13.5 g) was added slowly and the mixture stirred for 30 minutes. Acidification with concentrated hydrochloric acid caused precipitation of the product, which was filtered off, thoroughly washed with cold water and dried in a vacuum oven at 40° for 4 hours to give (20.4 g, 89%) of colourless needles which proved to be the pure acetyl derivative m.p. 208 - 209° (lit.[233] m.p. 208°).

N-Acetyl-3-acetoxyindole (25)

N-Acetylphenylglycine-o-carboxylic acid (10.0 g, 0.042 mol), was dissolved in acetic anhydride (50 cm^3) containing triethylamine (10 cm^3). The mixture was heated at reflux temperature under a protective nitrogen atmosphere for 20 minutes.

The acetic anhydride and triethylamine were then evaporated under a pressure of 0.3 mm at a temperature of 40° for 2 hours. This gave a sticky solid which was extracted with hot petroleum ether (b.p. 60 - 80°) (5 x 150 cm^3) portions. The petroleum ether extracts were combined and the volume reduced to (500 cm^3) by evaporation under reduced pressure. The extract was then heated at reflux temperature for 10 minutes with decolourising carbon (1 g). The solution was filtered hot into a chilled receiver and the product separated on cooling to give colourless needles (6.0 g, 65.6%), m.p. 80 - 81° (lit.[147] m.p. 80 - 81°). λ_{max} (ε) 234 (28,000), 264 (11,500), 293 (10,000), 301 (11,400) n.m. ν_{max} 1765 (C=O st for $OCOCH_3$), 1720 (C=O st for $NCOCH_3$), 1600 (Aryl C-H st), 1220 and 1180 (C-O st), 810, 750 (1,2 aromatic substitution) cm^{-1}. δ ($CDCl_3$) 2.30 (3H, s, $NCOCH_3$), 2.52 (3H, s, $OCOCH_3$), 7.2 - 7.6 (3H, m, 4, 5 and 6H), 7.62 (1H, s, 2H), 8.4 (1H, d x 2 $J_{7,6}$ 7Hz $J_{7,5}$ 2Hz - 7H). m/e 217 ($M^{+\cdot}$, 19%), 175 (31), 133 (100% base), 104 (9), 76 (43).

(E)- and (Z)-2-[1-(4-pyridyl)ethylidene] indolin-3-one (181)

To 4-acetylpyridine (3.32 g = 3.03 cm^3, 0.0275 mol or 1 molar equivalent) dissolved in deoxygenated methanol (50 cm^3) was added N-acetyl-3-acetoxyindole (25), (5.97 g, 0.0275 moles or 1 molar equivalent) and a solution of potassium hydroxide (16.6 g) in

deoxygenated distilled water (40 cm^3). The mixture was contained under a protective nitrogen atmosphere and stirring was continued for ten minutes, the flask was then sealed and set aside for seven days.

The reaction mixture was then poured rapidly into 20% aqueous acetic acid (500 cm^3) with constant stirring, maintaining the temperature at 5 - 10^o with the aid of an ice-bath. The acidic solution was extracted with dichloromethane in (4 x 300 cm^3) portions and the organic phases combined. The dichloromethane extract was washed thoroughly with 30% aqueous potassium carbonate solution in (5 x 100 cm^3) portions*. The dichloromethane extract was dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a mixture of (181 E)- and (181 Z)- as a dark-orange coloured solid (6.49 g, 88%) λ_{max} (ε) 270 (15,000), 485 (4,500) n.m. ν_{max} 3,100 (N-H st), 1700 (C=O st), 1640 (C=C st), 1620 (N-H def). m/e 236 ($M^{+\cdot}$, 100% base).

* If an emulsion forms at this point it may be broken by filtration through Kieselguhr.

† Methanol was deoxygenated by passing oxygen free nitrogen gas through the solution for 1 hour, prior to use, and the water was boiled for 45 minutes.

Separation of (E)- and (Z)-2- [1-(4-pyridyl) ethylidene] indolin-3-ones (181).

Interestingly, T.L.C. analysis of these products showed components on basic and neutral alumina and silica with a variety of solvents including: chloroform, chloroform/petroleum ether (b.p. 40 - 60^{o}) in a ratio of 3:2, dichloromethane and ethyl acetate/petroleum ether (b.p. 40 - 60^{o}) in a ratio of 9:1. All this work was carried out under conditions of normal illumination. However, if the T.L.C. analysis was performed in the absence of light the (E)- and (Z)-forms were seen to separate using a mixture of dichloromethane and methanol in a ratio of 98:2, with R_f values of 0.52 (E)- and 0.45 (Z)- using silica plates. Even in the dark other components were seen to form slowly or on prolonged elution and rapidly if the plates were exposed to light. For an explanation of this (see discussion section p. 144).

The mixed (E)- and (Z)-indoxylidines (181) were finally separated by the technique of 'flash' chromatography, as follows:

The mixed (E)- and (Z)-indoxylidines (181) (2.5 g) were dissolved in a mixture of dichloromethane and methanol in the ratio of 98:2 and chromatographed on a silica column in the absence of light. This was achieved by wrapping the column in aluminium foil under conditions of reduced illumination. The column was run rapidly under a positive nitrogen pressure of 2.1 Kg/cm^{2}, and under these conditions the E-isomer of (181) eluted close to the solvent front. The first band to emerge from the column was collected (approximately 150 cm^{3}) and the solvents evaporated under reduced

pressure to give green needles (0.3 g). T.L.C. analysis now showed this to be the pure E-isomer m.p. 210 - 215° (decomp.), λ_{max} (ε) 269 (15,100), 295 shoulder (12,400), 486 (4,400) n.m ν_{max} 3,125 (N-H st), 1695 (C=O st), 1640 (C=C st), 1620 (N-H def), 1600 (Aryl and pyridyl C-H st)cm^{-1}. ($CDCl_3$) 2.20 (3H, s, CH_3), 7.0 - 8.40 (6H, m, - indoxyl 4,5,6 and 7H and pyridyl 3 and 5H), 8.50 (1H, d x 2 $J_{6,5}$ 6Hz $J_{6,2}$ 2Hz - pyridyl 6H) 8 75 (1H, d x 2 J 6Hz and J 2Hz - pyridine 2H), 8.80 (1H, s, broad-exchanges with D_2O - NH). m/e 236($M^{+\cdot}$, 100% base), 221 (15), 207 (16), 122 (14), 106 (26), 77 (30)..

T.L.C. analysis of further column extracts showed a poor separation and the material remaining on the column formed a new series of yellow bands after a period of about two days, (see p. for a possible explanation of this).

(Ethyl)-triphenylphosphonium bromide

Triphenylphosphine (7.0 g, 0.0267 moles) was dissolved in dry benzene (50 cm^3) and a solution of bromoethane (4.36 g, 3.0 cm^3 = 0.0400 moles or 1.5 molar equivalents) also in dry benzene (20 cm^3) was added dropwise with stirring at 0 - 10° in an ice-bath. The mixture was stirred at this temperature for 20 minutes and then at room temperature for 3 hours, after which the reaction was left to stand overnight under anhydrous conditions. This caused the pure product to crystallize as white plates, which were collected washing with dry ether to give (9.71 g, 98%). As this salt is quite hygroscopic it was stored in a vacuum desiccator until required for use. m.p. 206 - 207° (lit. [234] m.p. 203 - 205°). ν_{max} 1600 (Aryl C-H st), 1500 (Aryl - P st), 1110 and 1000 (Quaternary Aryl - P), 930 and 900 (d), 780, 750, 720, 690 cm^{-1}. δ ($CDCl_3$) 1.4 (3H, heptet

J 18Hz - $\underline{CH}_3CH_2P$), 3.8 (2H, octet J 18Hz - CH_3 - $\underline{CH}_2P$), 7.8 (15H, m, aryl $\underline{H}^s$).

δ values represent the centres of the multiplets.

Attempted Wittig reaction of (E)- and (Z)-2-[1-(4-pyridyl) ethylidene]indolin-3-ones (181) with the ylide generated from (ethyl)-triphenylphosphonium bromide.

(Ethyl)-triphenylphosphonium bromide (6.28 g, 0.0170 mol) was dissolved in dry tetrahydrofuran (50 cm^3) under a protective atmosphere of dry, oxygen free nitrogen. n-Butyl lithium (10.6 cm^3 of a 1.6 M solution in hexane = 1.025 g or 0.0170 mol) were injected through a septum cap into the solution with stirring. Stirring was continued for 2 hours at room temperature and the colour of the solution changed from yellow to deep red. The (E)- and (Z)-indolin-3-ones (181) (4.0 g, 0.0170 mol) in dry T.H.F. (35 cm^3) were then added dropwise to the solution at 5 - 10^o in an ice-bath. After addition was complete the solution was warmed to room temperature and stirred for 4 hours under anhydrous conditions.

Water (30 cm^3) was then introduced carefully and the T.H.F. evaporated under reduced pressure. The residue was partitioned between water (100 cm^3) and dichloromethane in (4 x 50 cm^3) portions. The organic extracts were combined, dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a light-brown coloured solid.

The infrared spectrum of this solid showed that the carbonyl band at 1700 cm^{-1}, characteristic of the indoxylidine starting materials was now absent. However, the ^{1}H n.m.r. spectrum showed more aromatic protons than was consistent with the expected structures, indicating that the triphenyl group was still present. Working on the assumption that we might be dealing with an unexpectedly stable beatine we carried out the following experiment.

The residue from the previous experiment was dissolved in dry T.H.F. (50 cm^{3}) and heated at the reflux temperature for 8 hours. The T.H.F. was evaporated under reduced pressure and the residue again examined by infrared and ^{1}H n.m.r. techniques, but these gave results identical to those previously obtained.

It was clear from these results that the product was simply contaminated with triphenylphosphine oxide. In a bid to remove this contaminant we carried out the following chromatographic work.

1. The product (1.5 g) was dissolved in a mixture of ethyl acetate and petroleum ether (b.p. 40 - 60^{o}) in the ratio of 3:2 and chromatographed on a short column of basic alumina, eluting with the same solvent system. However, separation on the column was poor, and the products moved very slowly.

2. Hence we changed the support material to silica in an effort to reduce the tendency to bind to the column.With this support, the T.L.C. indicated that the optimum solvent combination was a mixture of dichloromethane, ether and petroleum ether (b.p. 40 - 60^{o}) in a

ratio of 3:1:1. On running a silica column with the same solvent system it was found that the components ran at a much faster rate than before but the separation of products observed on the T.L.C. plates was not reproduced on the column, and once again a contaminated product resulted.

Other solvent combinations that were seen to give some separation on silica T.L.C. plates were:

3.	Dichloromethane/methanol	98:2
4.	Chloroform/ether	3:2
5.	Chloroform/petroleum ether (b.p. 40 - 60°)	4:1

However, the Rf values for the separations were very close in all these solvent systems and extensive tailing of the bands was seen to occur. None of these T.L.C. results could be successfully transferred to a preparative scale column.

6. Preparative plates were then run at a loading of (50 mg) per plate with the dichloromethane/ether/petroleum ether (b.p. 40 - 60°) solvent combination mentioned previously, but a satisfactory separation could not be achieved.

This separation problem proved to be intractable, because any solvent combination sufficiently polar to elute the required products also eluted the triphenylphosphine oxide contaminant.

In a final bid to utilize the basicity of the pyridine nitrogen in the product to our advantage, we carried out the following experiment.

7. The product (1.0 g) was extracted with hot 2N-hydrochloric acid in (4 x 50 cm^3) portions. The acid extracts were combined and filtered while hot. The filtrate was cooled to 0 - 5° in an ice bath and saturated sodium hydrogen carbonate solution was added dropwise with constant stirring until the mixture was neutral to litmus. The product precipitated and was filtered off, washing with water (50 cm^3) followed by ether in (4 x 30 cm^3) portions. The resulting solid was dissolved in dichloromethane (75 cm^3) and the solution dried over anhydrous sodium sulphate. The solvent was then evaporated under reduced pressure. However, both T.L.C. and ^1H n.m.r. analysis of the product showed that the contaminant was still present.

At this stage we abandoned attempts to separate this mixture as we did not have an efficient high pressure liquid chromatograph available to use at this time. G.L.C. separation was also rejected because the products are very polar compounds with long retention times and doubtful thermal stabilities.

In a final bid to carry out this reaction sequence the following experiments were performed.

8. The mixture (1.0 g) was dissolved in ethanol (50 cm^3) and irradiated with ultraviolet light for 4 hours with constant stirring. The ethanol was then evaporated under reduced pressure and the residue examined by T.L.C. and ^1H n.m.r. techniques, but once again no ellipticine could be detected.

9. The mixture (0.6 g) was dissolved in dimethylsulphoxide (35 cm^3) and heated at 150° for 5 hours in a thermostatically controlled oil bath. The mixture was cooled to 30° and the

bulk of the D.M.S.O. evaporated on a rotatory evaporator under a pressure of 0.3 mm for $1\frac{1}{2}$ hours. The residue was partitioned between dichloromethane (50 cm^3) and water (50 cm^3). The dichloromethane extract was then washed with a further quantity of water in (4 x 20 cm^3) portions. The organic extract was then dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a light-brown coloured solid. However, T.L.C. and ^1H n.m.r. again proved that no ellipticine had formed.

2,4-Dinitrophenylacetic acid (214)

2-Nitrophenylacetic acid (25 g) in a round bottomed flask of (500 cm^3 capacity) was cooled to -10^o in an ice-salt bath. Fuming nitric acid (250 cm^3) was then added with caution and continuous mechanical stirring. The addition was very slow at first becoming more rapid later, and at such a rate that the temperature of the mixture did not rise above 15^o.

When addition was complete (approximately 30 minutes) the stirrer was withdrawn and the mixture heated at reflux temperature for 1 hour, cooled to $\sim 30^o$ and poured into ice-water (500 cm^3).

The product precipitated as a heavy white solid that was filtered off when cold, washing with a little ice-water. The near-pure product was crystallized from 25% aqueous ethanol, allowing the mother liquor to stand overnight, to give a pure white, microcrystalline solid, that after drying at 100^o for 5 hours in a vacuum oven yielded (20.3 g, 65%) m.p. 183 - 184^o

(decomp) (lit.[235] 179 - 180° (decomp). λ_{max} (ε) 203 (shoulder) (9,460), 237 (13,400), 350 (100) n.m. ν_{max} 3,100 - 2,550 (alkyl C-H and O-H st), 1700 (C=O st), 1600 (Aryl C-H st), 1350 and 1340 (d, Aryl -NO_2 antisymmetric st), 1365 (Aryl - NO_2 symmetric st), 1250 (C-O and O-H def (coupled), 1080, 920 (O-H def out of plane), 860 (Aryl - C-N st), 880 and 830 (1,2,4-aromatic substitution pattern) cm^{-1}. δ [$(CD_3)_2SO$] 4.15 (2H, s, $\underline{CH}_2$), 7.80 (1H, d, $\underline{J}_{6,5}$ 7Hz - 6$\underline{H}$), 8.48 (1H, d x 2 $\underline{J}_{5,6}$ 7Hz $\underline{J}_{5,3}$ 2Hz - 5$\underline{H}$), 8.70 (1H, d $\underline{J}_{3,5}$ 2Hz - 3$\underline{H}$). m/e 226 ($M^{+\cdot}$, 100%).

2-Nitro-4-aminophenylacetic acid (215)

A solution of sodium polysulphide was prepared by dissolving crystalline sodium sulphide ($Na_2S.9H_2O$) (32g) in water (120 cm^3) and adding finely powdered sulphur (8 g). The mixture was warmed until a clear solution was obtained. A mixture of 2,4-dinitrophenylacetic acid (20 g) and water (160 cm^3) was heated to gentle reflux under nitrogen with continuous mechanical stirring.

The sodium polysulphide solution was added dropwise to the refluxing and stirring solution over a period of 30 - 45 minutes. When addition was complete stirring and refluxing were continued for a further 20 minutes. The mixture was allowed to cool to room temperature and the solid filtered off, washing with a little cold water. The product was transferred to a mixture of concentrated hydrochloric acid (28 cm^3) and water (120 cm^3). The mixture was warmed to 80° and held at this temperature for 30 minutes with continuous mechanical stirring. The desired amine dissolved leaving sulphur and a small amount of unchanged starting material. The solution was filtered hot and the filtrate cooled to 10 - 15°

by the addition of crushed ice. Concentrated aqueous ammonia solution was then added dropwise, maintaining the temperature at 15 - 20° by the addition of ice whilst the pH was monitored on a meter. When the isoelectronic point was reached the product precipitated as an orange amorphous solid, which was filtered off, drained dry, and crystallized from the minimum quantity of hot methanol to give 2-nitro-4-aminophenylacetic acid as shining amber coloured needles, (9.93 g, 57.3%) m p. 186 - 187° (lit.[236] m.p. 184 - 186°. λ_{max} (ϵ) 203 (7,400), 233 (14,560), 370 (1,490) n.m. ν_{max} 3,450 and 3,400 (d, NH_2 st), 3,100 - 2,750 (alkyl C-H and O-H st), 1680 (C=O st and NH_2 def), 1600 (Aryl C-H st), 1540 (Aryl - NO_2 antisymmetric st), 1360 (Aryl - NO_2 symmetric st), 1310 (Aryl - NH_2 C-N st), 1250 (C-O st and O-H def (coupled), 1190, 1050, 950 (O-H def out of plane), 860 and 840 (Aryl C-N st and 1,2,4-aromatic substitution pattern) cm^{-1}. δ [$(CD_3)_2SO$] 3.60 (2H, s, $C\underline{H}_2$), 5.45 (2H, s, broad-exchanges with D_2O - $N\underline{H}_2$), 6.75 (1H, d x 2 $\underline{J}_{5,6}$ 7Hz $\underline{J}_{5,3}$ 2Hz - 5$\underline{H}$), 6.98 (1H, d $\underline{J}_{6,5}$ 7Hz - 6H), 7.05 (1H, d $\underline{J}_{3,5}$ 2Hz - 3$\underline{H}$), 7.20 (1H, s, broad-exchanges with D_2O - CO_2H). m/e 196 ($M^{+\cdot}$, 100%).

4-Methoxy-2-nitrophenylacetic acid (219)

A. Potassium salt of ethyl-4-methoxy-2-nitrophenylpyruvate (218)

Anhydrous ether (30 cm^3) was placed in a three necked, round bottom flask (500 cm^3 capacity), fitted with a dropping funnel (50 cm^3 capacity), nitrogen inlet, motor driven stirrer (with seal) and a reflux condenser protected with a calcium chloride guard tube.

Potassium (freshly cut into small pieces and degreased with petroleum ether (b.p. 40 - 60°) under a stream of dry nitrogen) (3.91 g = 0.10 g atom) was added quickly against a positive nitrogen pressure.

A slow stream of dry nitrogen was passed through the flask, above the surface of the stirred liquid, and a mixture of commercial absolute ethanol (25 cm^3) and anhydrous ether (20 cm^3) was added dropwise at such a rate as to maintain mild boiling. When all the potassium had dissolved (approximately two hours), the nitrogen was shut off and the solution allowed to cool to room temperature. When cool, anhydrous ether (250 cm^3) was added, followed by freshly distilled diethyl oxalate (14.6 g = 13.6 cm^3 = 0.1 mol.), with stirring. After ten minutes, a solution of 4-methoxy-2-nitrotoluene (16.7 g = 0.1 mol) in anhydrous ether (30 cm^3) was added dropwise over a period of five minutes. Stirring was continued for an additional ten minutes, after which the mixture was set aside to stand at room temperature for forty eight hours. During this time the deep-red potassium salt of ethyl-4-methoxy-2-nitrophenylpyruvate separated out. The salt was collected and washed with anhydrous ether until the filtrate remained colourless. The product was air dried to give a dark-red amorphous powder (17.93 g, 58.8%) m.p. > 300°, water soluble ν_{max} 1720 (C=O st), 1590 ($CO_2^{\ominus}$ st), 1520 (Aryl - NO_2 st), 1360 ($CO_2^{\ominus}$ st), 1080 and 1020 (C-O - OCH_3 st), 880 and 810 (1,2,4-aromatic substitution pattern), 750 (C-N def out of plane) cm^{-1}.

B. Oxidation to 4-methoxy-2-nitrophenylacetic acid (219).

The potassium salt of ethyl-4-methoxy-2-nitrophenylpyruvate (15 g = 0.049 mol) was dissolved in water (50 cm^3) and sodium hydroxide (0.6 g) was added. The alkaline solution was cooled to 10^o in an ice-bath and hydrogen peroxide (100 volume, 35 cm^3) added dropwise with stirring. Stirring was continued for a further two hours and the solution allowed to warm to room temperature.

The solution was then acidified with concentrated hydrochloric acid and the product precipitated at once. The solid was filtered off, washed with a little cold water, dissolved in dichloromethane and washed with saturated sodium hydrogen carbonate solution (3 x 20 cm^3). The organic layers were combined and dried over anhydrous sodium sulphate. The solvent was evaporated under reduced pressure to give the product as an almost pure, orange amorphous powder. Crystallization from methanol gave fine orange needles of 4-methoxy-2-nitrophenylacetic acid (6.55 g; 63.2%) m.p. 157 - 158° (lit. [83] m.p. 157 - 158°) λ_{max} (ε) 218 (13,760), shoulder at 240 (6,880), 269 (3,520), 345 (1,760) n.m. ν_{max} 3,300 - 2,500 (O-H st bonded), 1710 (C=O st), 1510 ($ArNO_2$ st), 1260 (antisymmetric stretch $OC\underline{H}_3$), 1040 (symmetric stretch OCH_3), 880 and 810 (1,2,4-aromatic substitution) cm^{-1}. δ [$(CD_3)_2SO$] 3,86 (3H, s, $OC\underline{H}_3$), 3.92 (2H, s, $C\underline{H}_2$), 7.25(1H, d x 2 $\underline{J}_{5,6}$ 8Hz $\underline{J}_{5,3}$ 4Hz - 5$\underline{H}$), 7.42 (1H, s, - 3$\underline{H}$), 7.55 (1H, d x 2 $\underline{J}_{6,5}$ 8Hz $\underline{J}_{6,3}$ 2Hz - 6$\underline{H}$), 11.50 (1H, s broad. Exchanges with D_2O - COO$\underline{H}$). m/e 211 ($M^{+\cdot}$, 41%), 166 (32), 150 (100, base), 135 (26), 122 (63), 108 (42), 77 (47), Metastable at 135.54 for (166 ⟶ 150).

6-Methoxyoxindole (220)

A mixture of 4-methoxy-2-nitrophenylacetic acid (5.0 g), 10% palladium carbon (500 mg) and glacial acetic acid (100 cm^3) was agitated under hydrogen at 50 atmospheres pressure, maintaining a temperature of 80° for 5 hours. Filtration of the catalyst and evaporation of the acetic acid under reduced pressure gave a straw coloured gum. This gum was dissolved in dichloromethane and washed with saturated sodium hydrogen carbonate followed by water until the washings were neutral to litmus. The organic phase was then dried over anhydrous sodium sulphate and evaporated under reduced pressure to give an off-white, amorphous solid. The material was almost pure, but was crystallized from methanol to give 6-methoxyoxindole as fine white needles m.p. 160 - 161° (lit.[164] 158 - 159°) (3.62 g, 94%), λ_{max} (ε) 258 (4,000) 286 (2,560), 294* (1,920) * inflextion n.m. ν_{max} 3,200 (N-H st associated), 1680 (C=O st), 1600 (Ar-H st), 1160, 1100 and 1040 (Ar-OCH_3 st), cm^{-1}. δ ($CDCl_3$) 3.45 (2H, s, $C\underline{H}_2$), 3.75 (3H, s, $OC\underline{H}_3$), 6.45 (1H, s, - 7H), 6.50 (1H, d x 2 $J_{5,4}$ 7Hz, $J_{5,7}$ 3Hz - 5H), 7.05 (1H, d $J_{4,5}$ 7Hz - 4H), 9.35 (1H, s, broad - NH). m/e 163 (100%, base),135 (60), 118 (14), 91 (5), 78 (10). Metastable at 110.15 for (163 ⟶ 135) - CO.

Attempted preparation of 6-methoxy-3-[1-(3-pyridyl)ethylidine] indoline-2-one (221)

3-Acetylpyridine (1.21 g, 1.1 cm^3, 0.010 mol) and 6-methoxyoxindole (1.62 g, 0.01 mol) were dissolved in dry benzene (25 cm^3) and pyrollidine (0.71 g, 0.83 cm^3) added. The mixture was heated under reflux for 6 hours, with a Dean-Stark trap on the condenser.

The dark brown solution was allowed to cool to 40° and the solvents evaporated under reduced pressure to give a sticky solid smelling strongly of 3-acetylpyridine. The solid was partitioned between 2M hydrochloric acid (50 cm^3) and dichloromethane (50 cm^3). The dichloromethane phase was separated, washed with saturated sodium hydrogen carbonate solution (2 x 50 cm^3), followed by water (2 x 50 cm^3). The dichloromethane was then dried over anhydrous sodium sulphate and evaporated under reduced pressure to give an off-white solid, this was crystallized from methanol to give white needles (1.52 g) m.p. 160 - 161°. The aqueous acidic layer was basified at 0 - 5° in an ice-bath with saturated sodium carbonate solution and extracted with ether (2 x 40 cm^3). The ether extracts were combined and dried over anhydrous sodium sulphate. Evaporation of the ether under reduced pressure gave a yellow liquid (0.8 g, 0.7 cm^3).

T.L.C. and mass spectral analysis of these isolated materials proved the white solid to be 6-methoxyoxindole and the liquid to be 3-acetylpyridine.

The experiment was repeated using larger quantities of pyrollidine and longer reaction times, but this gave rise to a high proportion of dark polymeric gums and oils in the isolated material and none of the desired product could be detected by T.L.C. or mass spectral analysis.

6-Methoxy-3-[1-(3-pyridyl)ethylidine]indolin-2-one (221)

A mixture of 6-methoxyoxindole (2.0 g, 0.012 mol) and 3-acetylpyridine (1.45 g, 0.012 mol) were dissolved in anhydrous methanol (that had previously been saturated with dry hydrogen chloride gas) (15 cm^3) and heated at gentle reflux for 7 hours.

On cooling to room temperature the hydrochloride of the product precipitated as an orange crystalline solid. The solid was filtered off and dissolved in the minimum quantity of hot water (25 cm^3). The solution was then made basic by the dropwise addition of saturated sodium hydrogen carbonate solution with vigorous stirring. When the mixture was just basic the product precipitated as a yellow amorphous solid. The solid was filtered off, washed with cold water, drained dry and crystallized from 95% aqueous ethanol to give a bright canary yellow, microcrystalline solid (2.42 g, 74%). m.p. 154 - 155°. λ_{max} (ε) 275 (20,500), 323 (6,600), 397 (2,100) n.m. ν_{max} 3,150 (N-H st), 1710 (C=O st), 1630 (C=C st), 1600 (Ar and py C-H st), 1120 and 1030 (Ar-OCH_3 st) cm^{-1}. δ ($CDCl_3$) 2.56 (3H, s, $C\underline{H}_3$), 3.80 (3H, s, $OC\underline{H}_3$), 6.50 (2H, complex d x 2 and s overlapping) $\underline{J}$ 8 and 2Hz - oxindole 5 and $7\underline{H}^s$ respectively) 7.2 - 7.7 (3H, m, - oxindole $4\underline{H}$ and the pyridine 4 and $5\underline{H}$), (8.45 (1H, d x 2 $\underline{J}_{6,5}$ 7Hz $\underline{J}_{6,4}$ 2Hz - pyridine $6\underline{H}$ and 8.50 (1H, d, $\underline{J}$ 2Hz - $2\underline{H}$ pyridine - signals overlap). 9.8 (1H, s broad, exchanges with D_2O - $N\underline{H}$). m/e 266 ($M^{+\cdot}$, 93%), 251 (100 base), 237 (5), 223 (9), 194 (4), 136 (6), 116 (5), 78 (5). Metastable at 236.84 for (266 ⟶ 251) - CH_3. (Found: C, 72.1; H, 5.3; N, 10.6; $C_{16}H_{14}N_2O_2$ requires: C, 72.2; H, 5.3; N, 10.5%).

6-Methoxy-3[1-(3-pyridyl)ethyl]indolin-2-one (223)

A solution of 6-methoxy-3[1-(3-pyridyl)ethylidine]indolin-2-one (2.0 g) in 95% aqueous ethanol (50 cm^3) was maintained at a temperature of 80° (just below its boiling point). Sodium borohydride was added in small portions to the hot solution with vigorous stirring over a period of 30 minutes. The reaction was judged complete when a constant ultraviolet spectrum was observed.

Distilled water (20 cm^3) was then added and the ethanol removed by evaporation under reduced pressure. The pale yellow slurry was diluted with distilled water (150 cm^3) and extracted with dichloromethane (5 x 40 cm^3) portions. The organic phases were combined and dried over anhydrous sodium sulphate. Removal of the solvent under reduced pressure gave the product as a colourless, almost pure oil. (1.94 g, 96.4%) λ_{max} (ε) 244 (10,900), 252 shoulder (8,500) n.m. ν_{max} 3,210 (N-H st), 1725 (C=O st), 1600 (Ar and py C-H st), 1120 and 1035 (Ar-OCH_3 st) cm^{-1}. We did not attempt to crystallize all of the product oil as past experience in this laboratory with a similar mixture of diastereoisomers had shown that it is a very time consuming procedure. The bulk of the oil was carried forward directly to the next stage of the synthesis. However, a small amount was triturated with chloroform/petroleum ether (40 - 60°) followed by crystallization from 95% aqueous ethanol to give a sample of white microcrystalline product m.p. 173 - 176°. δ ($CDCl_3$) 1.28 and 1.32 (3H, d J 7Hz - CH_3CH), 2.9 and 3.1 (2H, m, CH - CH - CH_3),

6.50 - 7.30 (5H, m, - pyridine 4 and 5H^{s} and oxindole 4,5 and 7 H^{s}), 7.9 (1H, d x 2 $J_{6,5}$ 8Hz $J_{6,4}$ 2Hz - 6H), 8.15 (1H, d J 2Hz - 2H), 9.20 (1H, s, broad exchanges with D_2O). m/e 268 ($M^{+\cdot}$, 62%), 253 (100 base) - 15 (CH_3).

2-Chloro-3-[1-(3-pyridyl)ethyl]-6-methoxyindole (225)

A mixture of 6-methoxy-3-[1-(3-pyridyl)ethyl]indolin-2-one (1.50 g, 5.6 mmol) and phosphoryl chloride (1.55 g, 10.2 mmol = 0.94 cm^3) was heated with stirring at 110° for 5 hours under a nitrogen atmosphere. After the reaction was complete the solution was cooled to 0 - 5° in an ice-bath and excess phosphoryl chloride was decomposed by adding saturated sodium carbonate solution dropwise with stirring. The basic mixture was then extracted with ethyl acetate (3 x 20 cm^3) portions. The combined organic phases were dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a pale yellow oil. The oil was chromatographed on a short column of silica gel (Merck- Kieselgel 60) (75 g), eluting with ethylacetate. Collection of the first band to leave the column and evaporation of the solvent under reduced pressure gave a white amorphous solid. Crystallization from ethyl acetate/petroleum ether (b.p. 60 - 80°) in the ratio of 3:2 gave small white prisms after several hours. m.p. 159 - 160° (1.06 g, 66%). λ_{max} (ε) 261 (11,450), 273 (9,600), 281 (6,900) n.m. ν_{max} 3,150 (N-H st), 1600 (Ar and py C-H st), 1120 and 1030 (Ar-OCH_3 st) and 740 (C-Cl st) cm^{-1}. δ [$(CD_3)_2SO$] 1.80 (3H, d, J 10Hz $CHCH_3$), 3.82 (3H, s, OCH_3), 4.50 (1H, q, J 10Hz $CHCH_3$), 6.90 (1H, d x 2 J 8Hz and J 2Hz - indole 5H), 7.0 - 7.5 (3H, m - indole 4 and 7H^{s} and pyridine 5H), 7.7 (1H, d x 2 J 7Hz and J 2Hz -

pyridine 4H), 8.20 (1H, d x 2 J 7Hz J 2Hz - 6H), 8.50 (1H, d J 2Hz - 2H), 11.55 (1H, s, broad exchanges with D_2O - NH). m/e (% Relative intensity). 289 (M + 2, 21), 287 ($M^{+\cdot}$ 64), 274 (34), 272 (100, base), 236 (17), 210 (14), 173 (7), 151 (5), 136 (7). Metastables at 257.87 for (287 ⟶ 272) corresponding to - 15 - CH_3), and 205.37 for (272 ⟶ 236) corresponding to (- 36 -HCl).

(Found: C, 66.8; H, 5.1; N, 9.8. $C_{16}H_{15}ClN_2O$ requires: C, 66.9; H, 5.2; N, 9.7%).

3-[1-(3-pyridyl)-ethyl]-6-methoxyindole (161) (Method B).

A mixture of 2-chloro-3-[1-(3-pyridyl)ethyl]-6-methoxyindole (750 mg, 2.61 mmol), and 10% palladium on carbon (750 mg) in ethanol (100 cm^3) with triethylamine (2.5 cm^3) was agitated under hydrogen at a pressure of 17.6 Kg/cm^2 at 60° for 16 hours. The mixture was cooled to room temperature, the catalyst filtered off and the solvent evaporated under reduced pressure to give a yellow oil. The oil contained traces of triethylamine, so it was maintained at 70° under a pressure of 3 mm for 4 hours. A sticky solid resulted which was dissolved in dichloromethane/ethyl acetate in a ratio of 1:1 and chromatographed on a short column of neutral alumina (50 g). The second band from the column was collected and the solvent evaporated under reduced pressure to give an off-white solid. The product was crystallized from the minimum amount of ethanol to give a white micro-crystalline solid. (371 mg, 56.4%) m.p. 167 - 168°.

The other physical data were identical with those for the Grignard product, quoted on (p. 282).

3-[1-(3-Pyridyl)ethyl]-6-methoxyindole, Optimized (Method C).

A solution of ethyl magnesium bromide was prepared in exactly the manner previously described for method A on (p. 281).

This solution was cooled to 0 - 5° in an ice-salt bath and 6-methoxyindole (5.0 g, 0.034 mol) in dry ether (30 cm^3) was added dropwise over a period of 20 minutes with stirring under nitrogen. At the end of this time a dense white suspension of 6-methoxyindolyl magnesium bromide precipitated. The suspension was dissolved by adding the minimum quantity of tetrahydrofuran (freshly distilled from lithium aluminium hydride) (25 cm^3) to the mixture, which formed a clear orange solution.

To this solution maintained at 0 - 5° was added 3-(1-chloroethyl) pyridine (4.81 g, 0.034 mol) in dry ether (10 cm^3), dropwise over ten minutes. Stirring was maintained at 5° for 4 hours followed by 1 hour at room temperature after which the mixture was set aside and left to stand at room temperature for a further 24 hours.

The solvents were then removed by evaporation under reduced pressure to give a sticky, dark orange gum. The gum was cooled to 0 - 5° in an ice-salt bath and dissolved in 2M hydrochloric acid (50 cm^3) with vigorous stirring. Ether (50 cm^3) was added and the layers separated. The aqueous layer was washed with ether (4 x 20 cm^3), and then adjusted to pH 8-9 with concentrated aqueous ammonia solution,

added dropwise at less than 20° in an ice-bath. The basic solution was extracted with ether (3 x 35 cm^3), followed by dichloromethane in (4 x 35 cm^3) portions. The dichloromethane extracts were combined, dried over anhydrous sodium sulphate and heated to reflux for 5 minutes with decolourising carbon (200 mg). The hot solution was filtered and the dichloromethane evaporated under reduced pressure to give a yellow oil. The oil was triturated with dry ether to give an off-white solid. This material was crystallized from 95% aqueous ethanol to give a white microcrystalline solid (3.43 g, 40%) m.p. 166 - 167°.

All the other spectral and analytical data were identical with those previously quoted for this product, from method A, on (p. 282).

N-Acetyl-3-[1-(3-pyridyl)ethyl]-6-methoxyindole (228).

3-[1-(3-Pyridyl)ethyl]-6-methoxyindole(3.0 g) was heated under reflux with acetic anhydride (20 cm^3) and triethylamine (4 cm^3) for 30 minutes. The solvents were removed at a temperature of 40° under a pressure of 4mm for 3 hours, to give a green gum. The gum was dissolved in dichloromethane (50 cm^3) and the organic solution washed with saturated sodium hydrogen carbonate solution in (4 x 20 cm^3) portions. The organic layer was then dried over anhydrous sodium sulphate and evaporated under reduced pressure to yield a buff coloured solid. This amorphous material was crystallized twice from 95% aqueous ethanol to give a white microcrystalline solid (3.05 g, 87.2%), m.p. 133 - 134° λ_{max} (ε) 244 (18,400), 257 (14,600), 276 shoulder (5,400), 280 (3,200),

316 (2,100) n.m. ν_{max} 1700 (C=O st), 1600 (Ar and py C-H st), 1170 and 1030 (Ar-OCH_3 st), 850, 810, 750 and 700 cm^{-1}.

δ [$(CD_3)_2SO$] 1.68 (3H, d J 6Hz $\underline{CH}_3CH$), 2.45 (3H, s, $NCO\underline{CH}_3$), 3.90 (3H, s, $O\underline{CH}_3$), 4.35 (1H, q, J 6Hz $\underline{CH}CH_3$), 6.80 - 7.30 (5H, m, indolyl-2,4,5 and $7\underline{H}^s$ and pyridine 5H), 7.55 (1H, dt, J 8Hz J 2Hz - pyridine 4H), 8.45 (1H, d x 2 J 8Hz J 2Hz - pyridine 6H), 8.55 (1H, d J 2Hz - pyridine 2H). m/e 294 ($M^{+\cdot}$, 40%), 279 (83), 264 (24), 252 (50), 237 (100, base), 222 (33), 212 (58), 174 (66), 162 (33), 135 (75), 114 (50), 106 (70), 78 (18).

(Found: C, 73.3; H, 6.0; N, 9.6 $C_{18}H_{18}N_2O_2$ requires: C, 73.5; H, 6.1; N, 9.5%).

Ethylacetimidate hydrochloride

Acetonitrile (105 g, dried by distillation over phosphorus pentoxide) was mixed with absolute ethyl alcohol (119 g, 1 molar equivalent) and anhydrous diethylether (170 cm^3). Dry hydrogen chloride gas was passed through the mixture, maintained under anhydrous conditions, and cooled in an ice-bath at 0 - 10°. When one molar equivalent of hydrogen chloride had been absorbed (indicated by a weight increase of 83 g), the solvents were removed by evaporation under reduced pressure. The white crystalline product was collected and washed with dry ether. This product is very hygroscopic, so it was quickly stored in a vacuum desiccator, over both potassium hydroxide (to remove any residual hydrogen chloride) and phosphorus pentoxide, the yield was (273.3 g, 86.4%) of white plates, m.p. > 300°, ν_{max} 1640 (C=N st).

1-Ethoxy-1-oximidoethane (42)

A solution of potassium carbonate (224 g, 2 molar equivalents) in water (500 cm^3) was prepared and cooled in an ice-bath to 5°, then ethyl acetimidate hydrochloride (100 g = 0.809 mole) was added all at once with vigorous stirring. The mixture was transferred to a separating funnel and shaken vigorously for ten minutes. The organic layer was then separated and the aqueous phase extracted with ether in (4 x 100 cm^3) portions. The organic extracts were combined and washed with water in (3 x 100 cm^3) portions. The combined ethereal layers were cooled in an ice-bath to 5° and a solution of hydroxylamine hydrochloride (60.4 g, 1.25 molar equivalents) in water (250 cm^3) was added with vigorous stirring. The mixture was transferred to a separating funnel and shaken vigorously for fifteen minutes. The ether layer was separated and the aqueous layer extracted with ether in (3 x 100 cm^3) portions. The ether layers were combined and dried over anhydrous sodium sulphate. The ether was then evaporated under reduced pressure to give the product as a colourless, viscous liquid, which had a strong and characteristic odour of roasted nuts. The yield was (43.4 g, 52%). Cooling the oily liquid in a refrigerator caused solidification to give white needles m.p. 19°. ν_{max} 3,600 - 3,200 (H-bonded O-H st), 1665 (C=N st), 1300 (O-H def in plane), 1050 (O-H def coupled with C-O def) cm^{-1}.

Mesitylene sulphonyl chloride

To chlorosulphonic acid (175 g = 100 cm^3 or 1.5 molar equivalents), stirred mechanically in an ice-bath at 0 - 5°, mesitylene (120 g = 140 cm^3 or 1 molar equivalent) was added cautiously, at such a rate that the temperature did not rise above

20^{o}. When addition was complete stirring was continued for one hour, and the reaction mixture poured, in a thin stream onto crushed ice (300 g) with constant stirring. The product was extracted with carbon tetrachloride in (3 x 400 cm^3) portions. The combined organic layers were washed with 30% aqueous sodium carbonate solution in (2 x 200 cm^3) portions, dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a colourless oil, which crystallized on cooling to room temperature. The product was collected and recrystallized from petroleum ether (b.p. 40 - 60^{o}) to give (175.6 g, 87.8%) of long white needles. m.p. 56 - 57^{o} (lit.[237] m.p. 56 - 57^{o} ν_{max} 1600 and 1580 (d - Ar - H st), 1360 (SO_2 asymmetric st), 1180 and 1170 (SO_2 symmetric st), 850, 780 and 740 (1,2,3,5-substitution on the aryl ring) cm^{-1}.

Ethyl-O-mesitylenesulphonylacetohydroxamate (43)

To dimethylformamide (150 cm^3) was added 1-ethoxy-1-oximido-ethane (40 g = 0.388 mole) and triethylamine (43.4 g = 1.1 molar equivalents). The mixture was stirred at 0 - 5^{o} in an ice-bath and mesitylene sulphonyl chloride (84.8 g = 1.0 molar equivalent) was added portion-wise over a period of thirty minutes, the temperature being maintained below 15^{o}. When addition was complete the solution was stirred for a further hour at 10 - 15^{o}, and poured onto crushed ice (800 g) which caused the separation of a white oil, vigorous agitation for five minutes effected solidification to a white solid. The product was filtered off on a sintered glass funnel, washing with petroleum ether (b.p. 60 - 80^{o}) drained dry and stored at once in a refrigerator at -20^{o}. The product was in a near pure state so further purification was not attempted because

past experience has shown that this compound was quite unstable at room temperature. The yield was (94.6 g, 85.6%), m.p. 48 - 50° with decomposition. ν_{max} 1645 (C=N st), 1360 (SO_2 - O - asymmetric st) and 1180 (SO_2 - O - symmetric st) cm^{-1}.

Notes:

1. The triethylamine concentration is critical in this preparation. Too little and incomplete reaction results. Too much and crystallization cannot be induced.
2. The temperature must be kept low or darkening will occur, giving rise to an impure product.

O-Mesitylene sulphonylhydroxylamine (M.S.H.)

To ethyl-O-mesitylenesulphonylacetohydroxamate (40 g = 0.140 mole) was added 70% perchloric acid (104 cm^3) with efficient stirring. The mixture was warmed to 30 - 35° on a water bath to aid solution and then stirred at room temperature for twenty minutes. The solution was then poured in a thin stream, into ice-water (600 cm^3), causing the product to precipitate, it was filtered off and transferred to ice-cold 1M sodium hydrogen carbonate solution (250 cm^3). After two minutes stirring the solid was again collected and washed with cold water. This product was dissolved in ether, and the ether layer separated from residual water. Finally the ether layer was dried over anhydrous sodium sulphate and evaporated under reduced pressure at not greater than 25°. The white microcrystalline product was immediately stored at -20° in a refrigerator since it is very unstable at room temperature. The yield was (20.7 g, 68.7%)

ν_{max} 3,270 and 3,230 (d-NH_2 st), 1365 (SO_2 st and SO_2 - O asymmetric st), and 1170 (SO_2 - O - symmetric st) cm^{-1}.

Note:

It is very important to keep the temperature of this product below 25°, above this temperature the material may take fire spontaneously.

1-Amino-3-[1-(N-acetyl-6-methoxyindolyl)ethyl] pyridinium mesitylenesulphonate (229).

N-Acetyl-3-[1-(3-pyridyl)ethyl]-6-methoxyindole (2.5 g) was dissolved in dichloromethane (40 cm^3) and cooled to 0 - 5° in an ice-bath. To this was added a solution of O-mesitylene sulphonylhydroxylamine (M.S.H.) (1.83 g, 1 molar equivalent) in ice-cold dichloromethane (30 cm^3), dropwise with stirring. Stirring was continued at room temperature for 25 minutes. The solution was then poured into anhydrous diethylether (500 cm^3) which caused immediate precipitation of a cream solid. The precipitate was allowed to settle for 30 minutes and the bulk of the ether was then removed by decantation. The material was collected on sintered glass and washed well with anhydrous ether. The salt was crystallized from the minimum amount of methanol to give a white microcrystalline solid (4.12 g, 95.2%) m.p. 182-184° ν_{max} 3,400 and 3,330 (d - NH_2 st), 1700 (C=O st), 1640 and 1620 (N-H def), 1600 (Aryl and pyridine C-H st), 1170 and 1030 (Ar-OCH_3 st) cm^{-1}. δ [$(CD_3)_2SO$] 1.70 (3H, d $\underline{J}$ 6Hz - $CHC\underline{H}_3$), 2.15 (3H, s, $C\underline{H}_3$), 2.50 (6H, s, - 2 x CH_3), 2.70 (3H, s, $NCOC\underline{H}_3$), 3.91 (3H, s, $OC\underline{H}_3$), 4.36 (1H, q, $\underline{J}$ 6Hz - $C\underline{H}CH_3$), 6.54 (2H, s, - mesitylate Ar-$\underline{H}$), 7.2 - 7.4 (4H, m, - indolyl 4,5 and 7$\underline{H}$, and pyridyl 5$\underline{H}$), 7.8 (1H, 2 x d x d $\underline{J}$ 8Hz and $\underline{J}$ 2Hz - pyridyl 4$\underline{H}$),

8.3 - 8.5 (3H, m, - indoyl 2H and NH_2 - exchanges with D_2O), 8.80 (1H, d x 2 $J_{6,5}$ 8Hz and $J_{6,2}$ 2Hz - pyridyl 6H), 9.15 (1H, d J 2Hz - pyridyl 2H).

3-[1-(N-Acetyl-6-methoxyindolyl)ethyl]pyridine N-acetylimide (230)

The product from the previous reaction (3.5 g) was dissolved in water (60 cm^3) and the solution cooled to 0 - 5° in an ice-bath. Acetic anhydride (70 cm^3) was added dropwise with continuous stirring, maintaining the temperature below 10° during the addition. The mixture was allowed to warm up to room temperature and stirred at this temperature for 30 minutes. The solution was then rapidly basified with 30% aqueous potassium carbonate solution (excess foaming was controlled by adding a trace of ether to the mixture), stirring vigorously and monitoring the pH on a meter. When just basic the solution was quickly extracted with dichloromethane in (4 x 100 cm^3) portions. The organic layers were combined, dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a pale amber oil which crystallized to give needles (2.36 g, 98%) m.p. 160 - 162° ν_{max} 1700 (N-C=O.CH_3 st), 1600 (Aryl and pyridyl C-H st), 1590 ($-\overset{\oplus}{N}$-C=O st), 1160 and 1035 (Ar - OCH_3 st)cm^{-1} δ [$(CD_3)_2SO$] 1.70 (3H, d J 6Hz $CHCH_3$), 2.10 (3H, s, $-\overset{\oplus}{N}$-CO-CH_3), 2.75 (3H, s, $NCOCH_3$), 3.90 (3H, s, OCH_3), 4.35 (1H, q, J 6Hz $CHCH_3$), 7.2 - 7.5 (4H, m, - indolyl 4,5 and 7H, and pyridyl 5H), 7.88 (1H, 2 x d x d J 8Hz and J 2Hz - pyridyl 4H), 8.42 (1H, s, - indolyl 2H), 8.92 (1H, d x 2 $J_{6,5}$ 8Hz and $J_{6,2}$ 2Hz - pyridyl 6H), 9.30 (1H, d, J 2Hz - pyridyl 2H).

1-(N-Methylacetamido)-3-[1-(N-acetyl-6-methoxyindolyl)ethyl] pyridinium iodide (231).

The product from the previous reaction (2.0 g) was treated with methyl iodide (30 cm^3) in acetone (60 cm^3) at reflux temperature for 45 minutes. The excess solvent and reagent were removed by evaporation under reduced pressure to afford a clear yellow oil. On cooling to room temperature the oil crystallized to form a pale yellow amorphous solid. The product was crystallized from the minimum amount of absolute ethanol at -20° overnight. The yield was (2.7 g, 96.4%) of pale yellow microneedles. m.p. 218 - 220° (dec) ν_{max} 1700 (N-C=O.CH_3 st), 1650 (- N(CH_3).C=OCH_3 st), 1600 (Aryl and pyridyl C-H st), 1150 and 1045 (Ar - OCH_3 st) cm^{-1}. δ [$(CD_3)_2SO$] 1.70 (3H, d J 6Hz - CHCH_3), 2.05 (3H, s, N(CH_3)COCH_3, 2.75 (3H, s, - NCOCH_3), 2.85 (3H, s, NCH_3), 3.90 (3H, s, OCH_3), 4.35 (1H, q, J 6Hz CHCH_3), 7.15 - 7.45 (4H, m, - indolyl 4,5 and 7H, and pyridyl 5H), 7.85 (1H, 2 x d x d J 8Hz and J 2Hz - pyridyl 4H), 8.40 (1H, s, - indolyl 2H), 8.90 (1H, d x 2 $J_{6,5}$ 8Hz and $J_{6,2}$ - pyridyl 6H), 9.30 (1H, d, J 2Hz - pyridyl 2H).

3-[1-(3-Pyridyl-4-carbonitrile)ethyl]-6-methoxyindole (232)

The methiodide salt from the previous reaction (2.0 g, 0.0041 mol) was dissolved in warm (25 - 30°) water (20 cm^3) by stirring for 5 minutes. This solution was treated with a mixture of ammonium chloride (0.56 g) and potassium cyanide (0.40 g = 0.0061 mol or 1.5 molar equivalents) dissolved in water (10 cm^3). The mixture was added dropwise over a period of 10 minutes at room temperature with stirring. When addition was complete stirring was continued for a further hour at room temperature,

during which time a tan coloured precipitate formed. The reaction mixture was extracted with chloroform in (4 x 40 cm^3) portions, and the organic layers combined. The chloroform solution was thoroughly washed with water in (4 x 30 cm^3) portions and then dried over anhydrous sodium sulphate. Evaporation under reduced pressure gave an amber coloured oil, which proved to be the expected 1,4-dihydrocyanopyridine intermediate.

ν_{max} 2,264 (C=N st), 1700 ($-NHCH_3$ $\underline{C=O}$ CH_3 st), 1680 ($N\underline{C=O}$ CH_3 st), 1640 (C=C st) cm^{-1}.

The oil was dissolved in ethanol and irradiated with 'soft' ultraviolet light for 30 minutes whilst stirring. The solvent was evaporated under reduced pressure to give a viscous orange coloured oil. The oil was dissolved in chloroform/methanol in the ratio of 98:2 and chromatographed on a short column of basic alumina, eluting with the same solvent system. The elute from the column was monitored by means of T.L.C. and ultraviolet light. The second band to emerge from the column was collected and left to stand over anhydrous sodium carbonate for 2 hours. The solvents were then evaporated under reduced pressure to give a straw coloured oil. Attempts to crystallize this oil proved very frustrating at first, but it was finally achieved from the minimum quantity of ethanol at -20° overnight, to give small white plates (878 mg, 78.2%). m.p. 92 - 93° λ_{max} (ε) 224 (28,400), 250 (4,400), 260 (6,350), 272 (4,000) 280 (2,500) n.m. ν_{max} 3,180 (N-H st), 2,260 (C=N st), 1625 (N-H def), 1600 (Aryl and pyridyl C-H st), 1160 and 1030 (Ar - OCH_3 st) cm^{-1}. δ [$(CD_3)_2SO$] 1.70 (3H, d $\underline{J}$ 6 Hz - $CH\underline{CH}_3$),

3.85 (3H, s, - $OC\underline{H}_3$), 4.35 (1H, q $\underline{J}$ 6Hz - $C\underline{H}CH_3$), 6.7 (2H, m, - indolyl 5 and 7$\underline{H}$), 7.25 (2H, m, - indolyl 2 and 4$\underline{H}$), 7.50 (1H, d x d $\underline{J}$ 6Hz and $\underline{J}$ 2Hz - pyridine 5$\underline{H}$) 8.20 (1H, d x d $\underline{J}_{6,2}$ 6Hz and $\underline{J}_{2,6}$ 2Hz - pyridine 6$\underline{H}$), 8.52 (1H, d $\underline{J}_{2,6}$ 2Hz - pyridine 2$\underline{H}$), 11.8 (1H, s-broad, exchanges with D_2O - N$\underline{H}$). $\underline{m/e}$ 277 ($M^{+\cdot}$, 84%), 262 (100, base), 250 (14), 237 (12), 199 (10), 159 (13), 131 (62), 77 (14).

(Found: C, 73.4; H, 5.4; N, 15.1; $C_{17}H_{15}N_3O$ requires: C, 73.6; H, 5.4; N, 15.2%).

Preparation of the Methyl lithium solution

The methyl lithium solution required for the conversion of the carbonitrile (232) to the iminoethyl compound (233) was prepared as follows:

Lithium (1.0 g, 0.143 mol) was cut into small pieces and degreased in petroleum ether (40 - 60^{o}) under a stream of nitrogen. The lithium was added to tetrahydrofuran (10 cm^3) (freshly distilled from L.A.H.) under anhydrous conditions. A solution of methyl iodide (5.0 cm^3 = 11.40 g = 1.1 molar equivalents) in dry T.H.F. (10 cm^3) was added to the stirring mixture warmed to 30^{o} in an oil-bath. One third of the mixture was added all at once to initiate the reaction, followed by the remainder at such a rate as to maintain a gentle reflux. After two hours all the lithium had dissolved, leaving traces of insoluble material. The solution was filtered under nitrogen, made up to (100 cm^3) with dry tetrahydrofuran and stored in a vacuum desiccator.

The molarity of the solution was determined by taking ($10\ cm^3$) aliquots and adding them to water ($50\ cm^3$). The resulting alkaline solution of lithium hydroxide was titrated with standard 0.1M hydrochloric acid using phenolphthalein as the indicator.

The volume of 0.1M hydrochloric acid consumed indicated the solution to be 0.64M.

3-[1-(3-Pyridyl-4-(1-iminoethyl)ethyl]-6-methoxyindole (233)

The carbonitrile (232) (600 mg), in dry T.H.F. ($25\ cm^3$) was added dropwise with stirring to a solution of methyl lithium ($10\ cm^3$ or 3 molar equivalents of the solution prepared previously) in anhydrous T.H.F. ($25\ cm^3$) at 0 - 5^o under an atmosphere of nitrogen. Stirring was continued for a further period of 30 minutes and then an ice-cold solution of ammonium chloride (0.60 g) in water ($30\ cm^3$) was introduced. The T.H.F. was evaporated under reduced pressure at less than 30^o, and the aqueous solution extracted with chloroform in ($4 \times 60\ cm^3$) portions. The combined organic layers were dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a colourless oil, attempts to crystallize this product at low temperature were not effective, but T.L.C., infrared and mass spectral data indicated it to be the desired compound in a near pure state, thus, it was hydrolysed directly in the next stage of the synthesis.

ν_{max} 3,240 (C=N-H st), 3,100 (N-H st), 1620 (N-H def), 1600 (Aryl and pyridyl C-H st), 1150 and 1040 ($Ar-OCH_3$ st) cm^{-1}.

m/e 293 ($M^{+\cdot}$ 31%), 278 (100, base).

8-Methoxyellipticine

3-[1-(3-Pyridyl-4-(1-iminoethyl))]-6-methoxyindole (400 mg) was dissolved in 20% aqueous acetic acid (150 cm^3) and heated on the steam bath at 85^o for 35 minutes. The solution was cooled to room temperature and basified with 30% aqueous potassium carbonate solution, dropwise with stirring. When basic to litmus the solution was extracted with ethyl acetate in (3 x 50 cm^3) portions. The organic layers were combined and dried over anhydrous sodium sulphate. The dry ethyl acetate solution displayed a strong yellow-green fluorescence under ultraviolet light. The solution was evaporated under reduced pressure to give a yellow solid. This amorphous material was crystallized carefully from the minimum amount of 95% aqueous ethanol to give a yellow microcrystalline solid (350 mg, 93%) m.p. 280 - 281° λ_{max} (ε) 248 (21,600), 280 (39,100), 300 (47,400), 310 shoulder (27,200), 340 (5,800), 360 (2,600) n.m. ν_{max} 3,180 (N-H st), 1605 (Ar-H st), 1270 (Ar-OCH_3 Antisymmetric st), 1150, 1050 and 1040 (Ar-OCH_3 symmetric st), 820, 810 cm^{-1}. δ [$(CD_3)_2SO$] 2.76 (3H, s, 11 $C\underline{H}_3$), 3.20 (3H, s, 5$C\underline{H}_3$), 3.92 (3H, s, $OC\underline{H}_3$), 7.25 (1H, d x 2 $\underline{J}_{9,10}$ 8Hz, $\underline{J}_{9,7}$ 2Hz - 9$\underline{H}$), 7.50 (1H, d $\underline{J}_{7,9}$ 2Hz - 7$\underline{H}$), 7.90 (1H, d, $\underline{J}_{10,9}$ 8Hz - 10$\underline{H}$), 8.35 (2H, s broad 3 and 4$\underline{H}^s$), 9.70 (1H, s, 1$\underline{H}$), 11.00 (1H, s broad exchanges with D_2O - N$\underline{H}$). m/e 276 ($\underline{m/e}^{+\cdot}$, 100% base), 261 (10), 246 (14), 231 (22), 149 (47), 123 (17), 110 (22), 97 (30), 85 (42), 71 (36). Metastable at 219.26 for 276 → 246 corresponding to $-CH_2O$

(Found: C, 78.2; H, 5.7; N, 10.1; $C_{18}H_{16}N_2O$ requires: C, 78.3; H, 5.8; N, 10.1%).

8-Hydroxyellipticine

A. Pyridinium chloride was prepared as follows:

Pyridine (freshly distilled from potassium hydroxide) (10 g) was mixed with annhydrous ether (50 cm^3) and saturated with dry hydrogen chloride gas until no further weight increase occurred. The white solid was filtered off under dry nitrogen and recrystallized from methanol to give colourless plates (14.5 g, 98%). This intensely hygroscopic material was stored at once, in a vacuum desiccator.

B. O-Demethylation of 8-methoxyellipticine

8-Methoxyellipticine (300 mg) was mixed with pyridinium chloride (3.5 g - prepared as in A above), and the temperature of the mixture raised to 215^o in a thermostatic oil bath. The mixture was held at this temperature for one hour, and then cooled to room temperature. Water (5 cm^3) was added followed by sufficient concentrated aqueous ammonia solution to adjust the mixture to pH 10. A dark solid resulted, which was allowed to settle, and the bulk of the solution was then decanted. The residue was extracted in boiling methanol to give a deep red solution which was dried over annhydrous sodium sulphate and evaporated under reduced pressure to give a brown solid. This material was chromatographed on preparative, silica gel T.L.C. plates using the solvent mixture quoted by Dat-Xuong: benzene/ethyl acetate/ethanol/concentrated aqueous ammonia solution in a ratio of 21:6:4:5. A band exhibiting a strong green fluorescence under ultraviolet light was observed.

This band was removed and extracted with hot methanol. The methanol extract was dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a yellow-brown solid. This amorphous product was crystallized carefully from the minimum quantity of methanol/ethyl acetate 1:1, to give a yellow microcrystalline solid (85 mg, 30%) m.p. 268 - 270° with decomposition (lit.[63] m.p. 268° with decomposition). A sample of 8-hydroxyellipticine, kindly supplied by Professor Rosazza allowed a mixed melting point to be determined. Mixed m.p. 268 - 270° not depressed. The T.L.C. data in three different solvent systems is shown in(Table 8).

Table 8

System	Rf	Literature Rf.
A. chloroform/ethanol/ glacial acetic acid 70:30:2 v/v	0.36	0.36
B. chloroform/methanol/ diethylamine 80:10:4 v/v	0.19	0.17
C. chloroform/methanol 4:1 v/v	0.34	0.35

λ_{max} (ε) 228 (32,400), 273 (37,200), 281 (41,700), 302 (72,400), 340 (4,270), 370 (3,240) n.m. ν_{max} 3,240 (O-H st), 1600(Ar C-H st), 1580, 1450, 1400 (O-H def in plane), 1160, 1110 and 1020 (O-H def out of plane), 960 and 805 cm^{-1}.

δ [$(CD_3)_2SO$] 2.75 (3H, s, 11$\underline{CH}_3$), 3.20 (3H, s, 5$\underline{CH}_3$), 6.70 (1H, d x 2 $\underline{J}_{9,10}$ 8Hz $\underline{J}_{9,7}$ 2Hz - 9$\underline{H}$), 6.94 (1H, d $\underline{J}_{7,9}$ 2Hz - 7$\underline{H}$), 7.85 (1H, d $\underline{J}_{3,4}$ 6Hz - 3$\underline{H}$), 8.15 (1H, d $\underline{J}_{10,9}$ 8Hz - 10$\underline{H}$), 8.40 (1H, d $\underline{J}_{4,3}$ 6Hz - 4$\underline{H}$), 9.75 (1H, s, -1$\underline{H}$), 11.14 (1H, s broad, exchanges with D_2O - N$\underline{H}$). m/e 262 ($m/e^{+\cdot}$, 100% base), 247 (42), 233 (34), 218(21), 204 (17), 190 (16), 178 (10), 176 (6), 164 (5), 163 (5), 151 (10).

(Found: C, 77.8; H, 5.4; N, 10.6; $C_{17}H_{14}N_2O$ requires: C, 77.9; H, 5.3; N, 10.7%).

1,4-Dimethylcarbazole (7)

Indole (40 g = 0.34 mol) and hexane-2,5-dione (38.7 g = 0.34 mol) were dissolved in absolute ethanol (170 cm^3) containing p-toluene sulphonic acid monohydrate (34 g = 0.18 mol). The solution was heated at reflux temperature for 3 hours, cooled and concentrated to low bulk under reduced pressure. The solution was diluted with diethyl ether (250 cm^3) and shaken with water (250 cm^3). The aqueous layer was separated and extracted with ether (3 x 200 cm^3). The combined organic layers were dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a dark red syrup. This was extracted with hot petroleum ether (b.p. 80 - 100°) (5 x 200 cm^3) and the resultant yellow solution heated at reflux temperature with decolourising carbon for ten minutes. The carbon was filtered off, and the almost colourless filtrate concentrated under reduced pressure; to give a mass of short white needles on cooling. The near pure product was recrystallized from petroleum ether (b.p. 80 - 100°) to give slender white needles

(38.6 g, 58%) m.p. 96 - 97° (lit.[12] m.p. 97 - 98°) ν_{max} 3390 (N-H st), 1610 and 1590 (Ar-H st), 810, 760 and 730 cm^{-1}.

δ ($CDCl_3$) 2.25 (3H, s, C_1 $\underline{CH}_3$), 2.80 (3H, s, C_4 $\underline{CH}_3$), 6.9 (1H, d $\underline{J}_{3,2}$ 7Hz - 3$\underline{H}$), 7.10 (1H, d $\underline{J}_{2,3}$ 7Hz - 2$\underline{H}$), 7.25 - 7.60 (3H, m, 6,7 and 8$\underline{H}^s$), 7.90 (1H, s-broad, exchanges with D_2O very slowly N-$\underline{H}$), 8.15 (1H, d $\underline{J}_{5,6}$ 7Hz $\underline{J}_{5,7}$ 2Hz - 5$\underline{H}$).

m/e 195 ($M^{+\cdot}$, 100% base), 180 (38), 166 (6), 152 (5), 144 (4), 99 (15).

3-Formyl-1,4-dimethylcarbazole (8)

Imidazole (6.98 g, 0.1025 moles or 1 molar equivalent), acetonitrile (100 cm^3) and trifluoroacetic anhydride (40 cm^3, 59.5 g, 0.283 mole or 2.76 molar equivalents) were mixed at room temperature and then heated to the reflux temperature. 1,4-Dimethylcarbazole (20 g, 0.1025 mole) in acetonitrile (75 cm^3) was added dropwise to the refluxing solution. The addition was carried out at such a rate, that refluxing was maintained. When addition was complete refluxing was continued for 4 hours, after which the mixture was cooled to 20° and the solvents evaporated under reduced pressure.

The residue was dissolved in (150 cm^3 batches), in a previously prepared mixture of ethanol (400 cm^3), water (200cm^3) and sodium hydroxide (20 g). The addition was carried out dropwise with cooling in an ice-bath and continual stirring The (150 cm^3) batches of solution were transferred to a three necked flask of (5 l capacity) equipped with a mechanical stirrer,

dropping funnel and reflux condenser. The reaction mixture was heated at reflux temperature for 20 minutes and then cooled to room temperature with stirring. The solution was diluted with water (2 l) and neutralized by the dropwise addition of concentrated hydrochloric acid to the stirring solution in an ice-bath.

The product precipitated, was collected, washed with cold water, and crystallized from ethanol to give (17.7 g) of white microcrystalline solid m.p. 215 - 216° (lit.[12] m.p.. 215 - 216°). The overall yield for the reaction is 77% compared with 50% for the more conventional Vilsmeier procedure, where the product requires extensive purification.

ν_{max} 3,230 (s, broad N-H st, associated), 1640 (C=O st, para - NH lowers frequency), and 730 cm^{-1}. δ ($CDCl_3$) 2.55 (3H, s, C_1 $\underline{CH}_3$), 3.10 (3H, s, C_4 $\underline{CH}_3$), 7.25 (1H, d x 2 $\underline{J}_{5,6}$ 7Hz and $\underline{J}_{5,7}$ 2Hz - 5$\underline{H}$), 7.3 - 7.6 (2H, m, 6 and 7$\underline{H}$), 7.65 (1H, s, 2$\underline{H}$), 8.10 (1H, d x 2 $\underline{J}_{8,7}$ 7Hz and $\underline{J}_{8,6}$ 2Hz - 8$\underline{H}$), 10.5 (1H, s, C$\underline{H}$O), 11.0 (1H, s-broad exchanges with D_2O N$\underline{H}$), m/e 223 (100%, base), 222 (64), 208 (3), 194 (44), 180 (7), 169 (14), A metastable ion at 168.7 for (223 ⟶ 194) corresponding to -CHO, 112 (9), 98 (10).

3-(2,2-Dimethoxyethyliminomethyl)-1,4-dimethylcarbazole (248).

To 3-formyl-1,4-dimethylcarbazole (11.8 g) was added 2,2-dimethoxyethylamine (8.5 g = 8.80 cm^3 or 1.53 molar equivalents), the reactants were mixed to a slurry. This was heated on a steam bath for 2½ hours under a short air condenser. The solution was allowed to cool, and benzene (50 cm^3) was added and the mixture evaporated to dryness under reduced pressure. This gave a buff coloured solid which was crystallized from benzene to yield (15.3 g, 94%) of white microcrystalline solid. m.p. 146 - 147° λ_{max} (ε) 238, (8,400), 240 sn (6,500), 282 (7,450), 314 (16,300), 388 (6,200) n.m. ν_{max} 3,310 (NH), 1630 (C=N), 1590, 1070 (OCH_3), 730 and 745 cm^{-1}. δ ($CDCl_3$) 2.40 (3H, s, C_1 $C\underline{H}_3$), 2.70 (3H, C_4 $C\underline{H}_3$), 3.45 (6H, s, 2 x $OC\underline{H}_3$), 3.85 (2H, d, $C\underline{H}_2CH$), 4.80 (1H, t, CH_2 $C\underline{H}$), 7.2 - 7.5 (3H, m, 6,7 and 8$\underline{H}$), 7.8 (1H, s, 2$\underline{H}$), 8.15 (1H, d, $\underline{J}$ 8Hz - 5$\underline{H}$), 8.55 (1H, sbr, N$\underline{H}$), 8.70 (1H, s, C$\underline{H}$=N). m/e 308 ($M^{+\cdot}$ 91%), 293 (1), 277 (21), 261 (16), 249.1 metastable for (308 ⟶ 277) - OCH_3, 245 (18), 233 (100, base), 218 (63), 206 (58), 190 (25), 182.1 metastable for (233 ⟶ 206), 75 (74).

3-(2,2-Dimethoxyethylaminomethyl)-1,4-dimethylcarbazole (249).

To 3-(2,2-dimethoxyethyliminomethyl)-1,4-dimethylcarbazole (15.00 g) in 95% aqueous ethanol (2 l) was added sodium borohydride (10.00 g), portionwise over a period of 10 minutes with stirring, maintaining the temperature below 15° by means of an ice-bath. The reaction was judged to be complete by the observation of a constant ultraviolet spectrum. The ethanol

was evaporated under reduced pressure and the residue partitioned between water (750 cm^3) and chloroform (2 l). The combined organic layers were dried over anhydrous sodium sulphate and evaporated under reduced pressure to give the crude amine, as a light yellow, highly viscous oil. Crystallization from 2:1 ethylacetate/hexane afforded (13.3 g, 88%) of fine white micro needles m.p. 130 - 131° (with decomp.) λ_{max} (ε) 248 (5,200), 320 (6,800), 342 (5,420) n.m. ν_{max} 3,400 (N-H - CH_2 st), 3,310 (N-H st), 1600 (Ar), 1030 and 1080 (Ar-OCH_3 st) cm^{-1}. δ ($CDCl_3$) 1.55 (1H, s broad exchanges with D_2O $CH_2\underline{NH}$), 2.40 (3H, s, C_1 $C\underline{H}_3$), 2.75 (3H, s, C_4 $C\underline{H}_3$), 2.85 (2H, d, $C\underline{H}_2CH$), 3.40 (6H, s, 2 x $OC\underline{H}_3$), 3.90 (2H, d broad, - $C\underline{H}_2$), 4.50 (1H, t, CH_2 $C\underline{H}$), 7.1 - 7.4 (3H, m, 6, 7 and 8$\underline{H}^s$), 7.6 (1H, s, - 2$\underline{H}$), 8.05 (1H, d x 2 $\underline{J}_{5,6}$ 8Hz $\underline{J}_{5,7}$ 2Hz - 5$\underline{H}$), 8.40 (1H, s broad, exchanges with D_2O slowly N-$\underline{H}$). m/e 311 ($M^{+\cdot}$, 2%), 236 (100, base).

3-N-Tosyl-(2,2-dimethoxyethylaminomethyl)-1,4-dimethylcarbazole (250).

To 3-(2,2-dimethoxyethylaminomethyl)-1,4-dimethylcarbazole (13.0 g) in tetrahydrofuran (100 cm^3) and water (70 cm^3) was added sodium carbonate (5.0 g) with stirring. p-Toluene sulphonyl chloride (7.5 g) was then added portionwise to the stirred mixture maintained at less than 10° in an ice-bath. When addition was complete the solution was allowed to warm to room temperature and the stirring continued for a further 2 hours. The reaction mixture was then poured into ice-water (500 cm^3) and the product precipitated at once and was filtered off, washing with petroleum ether (b.p. 60 - 80°). The crude

material was recrystallized from ethylacetate to give (18.6 g, 96%) of white crystalline plates. m.p. 210 - 211° λ_{max} (ε) 236 (5,600), 315 (10,500), 345 (4,400) n.m. ν_{max} 3,310 (N-H st), 1600 (Ar), 1165 (SO_2 st), 1060 and 1040 (Ar-OCH_3 st) cm^{-1}. δ [$(CD_3)_2SO$] 2.55 (3H, s, C_1 $\underline{CH}_3$), 2.65 (3H, s, $\underline{CH}_3PhSO_2$), 2.80 (3H, s, C_4 CH_3), 2.95 (6H, s, 2 x $O\underline{CH}_3$), 3.10 (2H, d, $\underline{CH}_2$ CH), 3.90 (1H, t, CH_2 $\underline{CH}$), 4.5 (2H, s broad, $\underline{CH}_2$), 7.0 (1H, s, 2-$\underline{H}$), 7.1 - 7.3 (3H, m, 6,7 and 8-$\underline{H}$), 7.5 (2H, d, $\underline{J}$ 8Hz) and 7.8 (2H, d, $\underline{J}$ 8Hz), CH_3-$PhSO_2$ $\underline{p}$-pattern), 8.1 (1H, d, $\underline{J}$ 8Hz - 5$\underline{H}$), 11.1 (1H, s broad, exchanges slowly with D_2O N-$\underline{H}$). $\underline{m/e}$ 466 ($M^{+\cdot}$, 42%), 433 (3), 403 (13), 376 (4), 310 (13), 279 (4), 248 (5), 235 (2), 208 (100, base), 193 (7).

5,11-dimethylpyrido[4,3-b]carbazole (Ellipticine)

To a solution of 3-N-tosyl-(2,2-dimethoxyethylaminomethyl)-1,4-dimethylcarbazole (5.0 g) in tetrahydrofuran (1.5 l) and water (200 cm^3) was added concentrated hydrochloric acid (100 cm^3), with thorough mixing. Stirring was continued for 15 minutes at 40° on a water bath. The solution became bright yellow in colour and exhibited a strong green fluorescence in ultraviolet light. Stirring was continued at room temperature for 3 days, the tetrahydrofuran was removed by evaporation under reduced pressure to give a heavy yellow precipitate of the hydrochloride. This was filtered off, dissolved in T.H.F. (350 cm^3) and washed with 30% aqueous potassium carbonate solution in (4 x 300 cm^3), portions until basic to litmus, followed by water (2 x 500 cm^3) and finally saturated sodium chloride solution (2 x 150 cm^3).

The T.H.F. was dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a brownish-yellow solid. This was chromatographed in two portions on a pair of short basic alumina columns, eluting with chloroform. The chloroform fraction containing the product ran well ahead of the very polar impurities, so this first band was collected. The chloroform fractions were combined and dried over anhydrous sodium sulphate. The dry chloroform was then evaporated under reduced pressure to give a yellow amorphous solid. This was crystallized from ethanol to give ellipticine (1.04 g, 39.3%) as yellow micro-needles m.p. 309 - 311°. (lit.[12] m.p. 309 - 313°). λ_{max} (ε) 238 (22,000), 275 (32,600), 285 (74,900), 294 (72,200), and 330 (4,500) n.m. ν_{max} 1600 (Ar-H st), 1280, 1030, 950, 800 cm^{-1}. δ [$(CD_3)_2SO$] 2.7 (3H, s, 11-$\underline{CH}_3$), 3.2 (3H, s, 5-$\underline{CH}_3$), 7.1 - 7.4 (3H, m, 7, 8 and 9 $\underline{H}^s$), 7.6 (1H, d x d $\underline{J}$ 8Hz and $\underline{J}$ 2Hz - 10$\underline{H}$), 8.4 (2H, s broad, 3 and 4$\underline{H}$), 9.7 (1H, d $\underline{J}$ 2Hz - 1$\underline{H}$), 11.5 (1H, s broad, N$\underline{H}$). $\underline{m/e}$ 246 ($M^{+\cdot}$, 100% base), 231 (36), 217 (5), 204 (4), 123 (41), 105 (32), 84 (82).

9-Bromoellipticine

A solution of ellipticine (500 mg = 0.0020 mol) in glacial acetic acid (30 cm^3) was prepared by stirring at room temperature for 5 minutes. To this was added, dropwise with stirring a solution of bromine (0.10 cm^3 = 0.0020 mol) in glacial acetic acid (2 cm^3). A yellow precipitate formed immediately and was filtered off. The precipitate was washed with 50% aqueous ammonia solution (concentrated ammonia/water 1:1)

followed by water, drained dry and recrystallized from benzene/ethanol 1:1, to give (615 mg, 93.2%) of yellow microneedles m.p. 318 - 319° (lit.[12] m.p. 318 - 319°) λ_{max} (ε) 250 (28,400), 271 (47,500), 279 (49,750), 287 (62,600), 300 (52,700) and 315 shoulder (5,600), 350 (4,600), 382 (3,800), 405 (3,700) n.m. ν_{max} 3,125 (N-H st), 1600 (Ar C-H st), 1270 1230, 1020, 780 and 600 (C-Br st) cm^{-1}. δ, 200 MHz [$(CD_3)_2SO$] 2.70 (3H, s, 11 $C\underline{H}_3$), 3.12 (3H, s, 5$C\underline{H}_3$), 7.45 (1H, d $\underline{J}_{7,8}$ 7Jz - 7$\underline{H}$), 7.63 (1H, d x 2 $\underline{J}_{8,7}$ 7Hz $\underline{J}_{8,10}$ 2Hz - 8$\underline{H}$). 8.27 (1H, d $\underline{J}_{10,8}$ 2Hz - 10$\underline{H}$), 8.31 (1H, d $\underline{J}_{4,3}$ 6Hz - 4$\underline{H}$), 8.40 (1H, d $\underline{J}_{3,4}$ 6Hz - 3$\underline{H}$), 9.87 (1H, s, -1$\underline{H}$), 12.11 (1H, s, broad, exchanges with D_2O -N$\underline{H}$). $\underline{m/e}$ 327/325 (93%, $m/e^{+\cdot}$ 100, base), 312 /310 (9 and 10), 245 (71), 228 (25), 216 (2), 91 (79). Metastable at 295.6 for (325 ⟶ 310) corresponding to loss of CH_3, and metastable at 184.7 for (325 ⟶ 245) corresponding to loss of bromine. (Found: C, 62.6; H, 4.0; N, 8.6; $C_{17}H_{13}N_2Br$ requires: C, 62.7; H, 4.0; N, 8.6%).

9-Nitroellipticine

A solution of ellipticine (500 mg, 0.0020 mol) in glacial acetic acid (50 cm^3) was prepared by stirring at room temperature for 5 minutes. A mixture of glacial acetic acid (10 cm^3) and concentrated nitric acid (0.14 cm^3 = 0.138 g HNO_3, 0.0022 mol) was added dropwise with stirring over a period of 5 minutes. When addition was complete concentrated sulphuric acid (0.32 cm^3 = 0.6 g, 0.006 mol) was added dropwise with stirring. Stirring was continued at room temperature for 15 minutes and then the solution was set aside and left to stand. After 2 hours a heavy yellow precipitate settled out.

The solid was collected and the filtrate retained*. The solid was redissolved in 6M-hydrochloric acid (10 cm^3) and reprecipitated by the dropwise addition of saturated sodium hydrogen carbonate solution at 0 - 15° in an ice-bath with continuous stirring until basic. The yellow precipitate was filtered off and crystallized from glacial acetic acid to give a yellow microcrystalline solid (306 mg, 52%). m.p. 357 - 359° with decomposition (lit.[12, 191] m.p. 350° with decomposition) λ_{max} (ε) 235 (22,000), 275 (32,500), 296 (72,400), 305 (68,600), 378 (14,600) n.m. ν_{max} 3,220 (N-H st), 1600 (ArC-H st), 1545 ($Ar-NO_2$ Antisymmetric st), 1280, 1030, 950, 840 ($C-NO_2$ symmetric st), 800 cm^{-1}. δ, 200 MHz [$(CD_3)_2SO$] 2.48 (3H, s, 11$\underline{CH}_3$), 3.20 (3H, s, 5$\underline{CH}_3$), 7.60 (1H, d $\underline{J}_{7,8}$ 8Hz - 7$\underline{H}$), 8.12 (1H, d $\underline{J}_{10,8}$ 2Hz - 10$\underline{H}$), 8.44 (2H, m, 3 and 4$\underline{H}^s$), 8.50 (1H, d x 2 $\underline{J}_{8,7}$ 8Hz $\underline{J}_{8,10}$ 2Hz - 8$\underline{H}$), 9.12 (1H, d $\underline{J}$ 2Hz - 1$\underline{H}$), 12.53 (1H, s, broad, exchanges with D_2O - N$\underline{H}$). $\underline{m/e}$ 291 ($M^{+\cdot}$, 100% base), 276 (3), 275 (3), 261 (34), 245 (61), 229 (13), 217 (8), 204 (4), 123 (6), 105 (7), 79 (4). Metastable at 206.27 for (291 ⟶ 245) corresponding to the simple fission $-NO_2$.

(Found: C, 70.1; H, 4.5; N, 14.3; $C_{17}H_{13}N_3O_2$ requires: C, 70.1; H, 4.5; N, 14.4%).

* The filtrate on basification and work up, was found to contain (212 mg, 42.4%) of unchanged ellipticine.

3-(2-Methyl-1,3-dioxolan-2-yl)pyridine (262)

3-Acetylpyridine (55 g = 50 cm^3 = 0.45 moles) was heated with a solution of p-toluene sulphonic acid (85.2 g = 0.495 moles or 1.1 molar equivalents) in ethylene glycol (125 cm^3) and dry benzene (500 cm^3) with continuous removal of water in a closed trap, Dean Stark apparatus for 16 hours.

The mixture was cooled to room temperature, separated, and the lower ethylene glycol layer diluted with ice-water (500 cm^3). This solution was basified using solid sodium carbonate, portionwise with constant stirring. The basic solution was extracted with dichloromethane (4 x 400 cm^3) and the organic layers combined, dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a pale amber oil (74.5 g).

This experiment was repeated, and the products combined to give 148.5 g of amber oil. The product was purified by distillation at 110° under a pressure of 3.5 mm to give (128.5 g, 85.6%) of a colourless oily liquid. λ_{max} (ε) 275 (7,200)n.m. ν_{max} 2,990 and 2,880 (alkyl st), 1600 and 1590 (d- aryl C-H st), 1260, 1200, 1090 and 1040 (R-O-R st and def), 800, 880 and 710 (aromatic substitution (3 adjacent H^s and 1 lone H) cm^{-1} δ ($CDCl_3$) 2.68 (3H, s, CH_3), 3.80 and 4.05 (2H, heptets CH_2^s), 7.30 (1H, 2 x d (overlapping) $J_{5,4}$ 8Hz $J_{5,6}$ 8Hz - 5H), 7.80 (1H, d x 2 $J_{4,5}$ 8Hz $J_{4,2}$ 2Hz - 4H), 8.55 (1H, d x 2 $J_{6,5}$ 8Hz $J_{6,4}$ 2Hz - 6H), 8.75 (1H, d $J_{2,4}$ 2Hz - 2H). m/e 165 ($M^{+\cdot}$ 2%), 150 (100, base), 134 (10), 106 (58), 87 (30), 78 (30).

1-Amino-3-(2-methyl-1,3-dioxolan-2-yl)pyridinium Mesitylenesulphonate (263).

3-(2-Methyl-1,3-dioxolan-2-yl)pyridine (30.0 g = 0.18 moles) was dissolved in dichloromethane (100 cm^3) and cooled to 0 - 5° in an ice-bath. To this was added a solution of O-mesitylene sulphonylhydroxylamine (M.S.H.) (43 g = 1.1 molar equivalents) in ice cold dichloromethane (80 cm^3), dropwise with stirring. Stirring was continued at room temperature for 30 minutes. The solution was then poured into anhydrous diethylether (1.6 l) and again cooled to 0° and allowed to stand for an additional 30 minutes. During this time the product became crystalline and was collected, washing with anhydrous ether. The salt was recrystallized from the minimum amount of methanol to give a microcrystalline white solid (65.5 g, 94.8%), m.p. 118 - 119° λ_{max} (ε) 210 (5,760), shoulder 235 (1,500), 265 (960) n.m. ν_{max} 3,120 and 3,130 (NH_2 st), 1610 (NH def), 1600 (Ar C-H st), 1190 (SO_2 st), 1040 and 1010 (C - O - C st), 880, 840, 800, 740 (aromatic substitution) cm^{-1}. δ [$(CD_3)_2SO$] 1.60 (3H, s, -CC$\underline{H}_3$OCH_2CH_2O), 2.15 (3H, s, $\underline{p}$-C$\underline{H}_3$Ph$(CH_3)_2SO_3$), 2.50 (6H, s, 2 x CH_3 - CH_3 - Ph(C$\underline{H}_3)_2SO_3$), 3.75 and 4.0 (2H, heptets C$\underline{H}_2^s$), 6.70 (2H, s, benzenoid $\underline{H}^s$), 8.0 (1H, d x 2 $\underline{J}_{3,4}$ 7Hz $\underline{J}_{5,6}$ 7Hz - 5$\underline{H}$), 8.2 (1H, d x 2 $\underline{J}_{4,5}$ 7Hz $\underline{J}_{4,2}$ 2Hz - 4$\underline{H}$), 8.5 (2H, s broad NH_2 exchanges with D_2O), 8.70 (1H, d x 2 $\underline{J}_{6,5}$ 7Hz $\underline{J}_{6,4}$ 2Hz - 6$\underline{H}$) 8.75 (1H, d $\underline{J}_{2,4}$ 2Hz - 2$\underline{H}$).

3-(2-Methyl-1,3-dioxolan-2-yl)pyridine N-Acetylimide (264).

The product from the previous reaction (30 g = 0.079 moles) was dissolved in water (150 cm^3) and cooled to 5° in an ice-bath. Acetic anhydride (200 cm^3) (previously cooled to 5°) was added dropwise with continuous stirring, maintaining the temperature below 10° during the addition. The mixture was stirred rapidly at 5° for ten minutes and then basified by the dropwise addition of aqueous 30% sodium hydroxide solution (150 cm^3) followed by an aqueous solution of potassium carbonate (100 g in 900 cm^3 water). The base was added as rapidly as possible maintaining the temperature below 15° with constant stirring. When the solution was just basic it was rapidly extracted with dichloromethane (3 x 200 cm^3). The organic layers were combined and dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a straw-coloured oil (16.8 g, 96%) λ_{max} (ε) 270 (9,300), 318 (6,360) n.m. ν_{max} 1600 (Ar C-H st) 1580 (N-C=O), 880, 820 and 690 (aromatic substitution) cm^{-1}. δ ($CDCl_3$) 1.60 (3H, s, CH_3), 2.30 (3H, s, CH_3-C=O), 3.85 and 4.10 (2H, heptets CH_2 s), 8.40 (1H, 2 x d (overlapping) $J_{5,4}$ 8Hz $J_{5,6}$ 8Hz - 5H), 8.85 (1H, d x 2 $J_{4,5}$ 8Hz $J_{4,2}$ 2Hz - 4H), 8.70 (1H, d x 2 $J_{6,5}$ 8Hz $J_{6,4}$ 2Hz - 6H), 9.0 (1H, d $J_{2,4}$ 2Hz - 2H). m/e 222 ($M^{+\cdot}$ 3%), 207 (100, base).

No attempts were made to crystallize this compound due to its potentially unstable nature, previous attempts had indicated that long periods at -20° were required, so it was used immediately in the next stage of the synthesis.

1-(N-Methylacetamido)-3-(2-methyl-1,3-dioxolan-2-yl)-pyridinium iodide (265).

The pyridine (264) from the previous reaction (15.0 g, 0.0675 moles), was treated with methyl iodide (75 cm^3), at reflux temperature in acetone (100 cm^3) for forty five minutes. The excess reagent and solvent were removed by evaporation under reduced pressure to afford a clear, bright yellow oil. On cooling to room temperature this oil crystallized to form a mass of yellow needles. The product was recrystallized from the minimum quantity of hot ethanol to give (23.6 g, 96%) of stout shining yellow needles. m.p. 176 - 177° (lit.[18] m.p. 176 - 177°), λ_{max} (ε) 220 (19,400), 273 (5,900) n.m. ν_{max} 3000 - 2,850 (alkyl C-H st), 1650 (CH_3N - $\underline{C{=}O}CH_3$, 1600 (Aryl C-H st), (1110, 1060 and 1020 - C-O-C st and def), 870 850 and 810 cm^{-1}. δ [$(CD_3)_2SO$] 1.7 (3H, s, C-$\underline{CH}_3$), 2.5 (3H, s, $CH_3NC{=}O\underline{CH}_3$), 2.8 (3H, s, $NC\underline{H}_3$), 3.90 and 4.10 (2 x 1H, heptet's - O - $C\underline{H}_2$ - $C\underline{H}_2$ - O), 8.40 (1H, 2 x d(overlapping) $\underline{J}_{5,4}$ 8Hz $\underline{J}_{5,6}$ 8Hz - 5$\underline{H}$), 8.85 (1H, d x 2 $\underline{J}_{4,5}$ 8Hz $\underline{J}_{4,2}$ 2Hz - 4$\underline{H}$), 9.10 (1H, d x 2 $\underline{J}_{6,5}$ 8Hz $\underline{J}_{6,4}$ 2Hz - 6$\underline{H}$), 9.40 (1H, d $\underline{J}$ 2Hz - 2$\underline{H}$).

Attempted preparation of 3-(2-Methyl-1,3-dioxolan-2-yl)-4-ethylpyridine (266).

(3-(2-Methyl-1,3-dioxolan-2-yl)-6-ethylpyridine (273)

Magnesium turnings (1.34 g, 0.0549) moles or 2 molar equivalents) were covered with dry tetrahydrofuran (freshly distilled from L.A.H.) (25 cm^3) under a protective nitrogen atmosphere. Anhydrous conditions were maintained throughout and bromoethane (4.3 cm^3, 5.98 g or 2 molar equivalents), in dry T.H.F. (40 cm^3) was added with mechanical stirring, (15 cm^3) all at once, to initiate the reaction, followed by the remainder at such a rate as to maintain a gentle reflux. When the addition was complete, approximately 45 minutes, all the magnesium had dissolved leaving a clear grey solution of ethylmagnesium bromide.

Next, the pyridinium salt (265) from the previous reaction (10.0 g, 0.0274 moles) was dispersed in dry T.H.F. (50 cm^3) and the solution of ethylmagnesium bromide prepared previously added to it, with constant stirring under a protective nitrogen atmosphere. The addition was carried out dropwise with care, because the reaction proved to be quite exothermic. The mixture became chocolate-brown in colour and stirring was continued at room temperature for 4 hours, followed by heating at the reflux temperature for 16 hours under nitrogen.

The mixture was cooled under nitrogen and a 2% aqueous solution of ammonium chloride (100 cm^3) was introduced

to destroy any excess Grignard reagent. The T.H.F. was then removed by evaporation under reduced pressure and the resulting gum extracted with dichloromethane in (5 x 200 cm^3) portions. The dichloromethane phases were combined, dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a clear oil. The oil was dissolved in ethanol (50 cm^3) and irradiated with ultraviolet light for 3 hours with constant stirring. The ethanol was removed by evaporation under reduced pressure to yield the product as a clear, orange-coloured liquid (2.86 g, 54%).

T.L.C. analysis showed it to be a new compound: Rf values using neutral alumina plates and eluting with a mixture of dichloromethane/petroleum ether (b.p. 40 - 60°) in a ratio of 4:1 were as follows: Starting material 0.05 and product 0.94. λ_{max} (ε) 217 (6,500), 262 (650), 267 shoulder (2,600), 274 (7,200) n.m. ν_{max} 3000 - 2,850 (alkyl C-H st), 1600 and 1590 (d-aryl C-H st), 1480, 1370, (1260, 1200, 1040 and 1020 - R-O-R st and def), 950, (870, 840 and 800 - aromatic substitution pattern 1 lone H^s and 2 adjacent H^s) cm^{-1}. δ ($CDCl_3$) 1.25 (3H, t, $\underline{J}$ 8Hz - $C\underline{H}_3$ - CH_2), 1.62 (3H, s, $C\underline{H}_3$ - C), 2.84 (2H, q, $\underline{J}$ 8Hz - $C\underline{H}_2 \cdot CH_3$) 3.90 and 4.15 (2 x 2H, heptets - O - $C\underline{H}_2$ - $C\underline{H}_2$ - O), 7.12 (1H, d $\underline{J}_{5,4}$ 8Hz - 5$\underline{H}$), 7.68 (1H, d x 2 $\underline{J}_{4,5}$ 8Hz and $\underline{J}_{4,2}$ 2Hz - 4$\underline{H}$), 8.62 (1H, d $\underline{J}_{2,4}$ 2Hz - 2$\underline{H}$). $\underline{m/e}$ 193 ($M^{+\cdot}$, 5%), 178 (100, base).

From a consideration of the ^1H n.m.r. data (see discussion section p. 216) we concluded that the product of this reaction

was 3-(2-Methyl-1,3-dioxolan-2-yl)-6-ethylpyridine (273) rather than the desired -4-ethyl isomer.

We carried out the following experiments to try to overcome this difficulty.

1. The experiment was carried out as previously described but instead of refluxing the solution it was allowed to stand at room temperature for 24 hours. However, on working up this experiment the results were found to be identical with those obtained previously.

2. We considered the possibility that a photo chemical rearrangement or migration of the ethyl group might be occurring, so we attempted to aromatize the 1,4-dihydropyridine intermediate thermally, in accordance with Katritzky and Sammes[192] published method for the N-(2,6-dimethyl-4-oxopyridin-1-yl) pyridinium salts, that these authors employed. However, in our case it was found that heating the oil obtained before irradiation to 200^{o} for as short a time as ten minutes, resulted in an intractable polymeric product, presumably due to oxidative breakdown of the intermediate.

3-(2-Methyl-1,3-dioxolan-2-yl)pyridine-4-carbonitrile (275)

The methiodide salt (265) (5.0 g, 0.014 mol) was dissolved in warm water (30^{o}, 50 cm^{3}) with stirring. This solution was treated with a mixture of ammonium chloride (1.42 g) and potassium cyanide (1.02 g, 0.0208 mole or 1.5 molar equivalents),

dissolved in water (25 cm^3). The mixture was added dropwise over a period of 10 mimutes at room temperature with stirring. When addition was complete stirring was continued for a further hour at room temperature, during which time the solution turned deep orange in colour.

The reaction mixture was extracted with chloroform in (4 x 100 cm^3) portions, and the organic layers combined. The chloroform solution was thoroughly washed with water in (4 x 75 cm^3) portions and then dried over anhydrous sodium sulphate. Evaporation of the solvent under reduced pressure gave a light amber coloured oil, which proved to be the expected 1,4-dihydrocyanopyridine intermediate.

ν_{max} 2,265 (C≡N st), 1700 (-NCH_3 $\underline{C{=}O}$ CH_3 st), 1680 (N $\underline{C{=}O}$ CH_3 st), 1620 ($\underline{C{=}C}$ st) cm^{-1}

The oil was dissolved in ethanol and irradiated with 'soft' ultraviolet light for 30 minutes whilst stirring. The solvent was then evaporated under reduced pressure to give a viscous orange coloured oil. The oil was dissolved in diethyl ether and chromatographed on a short column of basic alumina eluting with ether. The progress of the column was monitored by ultraviolet light and T.L.C. The first band to emerge was collected and gave a strong blue fluorescence. The solvent was dried over anhydrous sodium sulphate and evaporated under reduced pressure to give a colourless oil which slowly crystallized

to afford white needles. (2.14 g, 82%) m.p. 68 - 69°
(lit.[18] m.p. 68 - 69°) λ_{max} (ε) 217 (7,850), 222 (6,700),
275 (3,900) n.m. ν_{max} 3000 - 2850 (alkyl C-H st), 2,220
(C=N st), 1600 (aryl C-H st), (1260, 1200, 1100 and 1040
-R - O - R st and def), 950, 890, 870, 850 and 800 cm^{-1}.
δ [$(CD_3)_2SO$] 1.70 (3H, s, $C\underline{H}_3C$), 3.90 and 4.05 (2 x 2H,
heptet's O-$C\underline{H}_2$-$C\underline{H}_2$-O), 7.8 (1H, d $\underline{J}_{5,6}$ 7Hz - 5$\underline{H}$), 8.7 (1H,
d x 2 $\underline{J}_{6,5}$ 7Hz $\underline{J}_{6,2}$ 2Hz - 6$\underline{H}$), 8.8 (1H, d $\underline{J}_{2,6}$ 2Hz - 2$\underline{H}$).
m/e 190 ($M^{+\cdot}$, 4%), 175 (100, base).

REFERENCES

References

1. S. Goodwin, A.F. Smith and E.C. Horning, J. Amer. Chem. Soc., 1959, 81, 1903.

2. G. Bucni, D.W. Mayo and F.A. Hochstein, Tetrahedron, 1961, 15, 167, and references cited therein.

3. J. Schmutz and F. Hunzicker, Helv. Chim. Acta, 1958, 41, 288.

4. R.B. Woodward, G.A. Jacobucci and F.A. Hochstein, J. Amer. Chem. Soc., 1959, 81,4434.

5. J.P. Wibaut and J.F. Arens, Rec. Trav. chim. 1941, 60, 119.

6. J. Bergman and R. Carlsson, J. Heterocyclic Chem., 1972, 9, 833.

7. B. Webb, Ph.D Thesis, University of Bath, 1974, 70.

8. P.A. Cranwell and J.E. Saxton, Chem. and Ind., 1962, 45, and J. Chem. Soc., 1962, 3482.

9. T.R. Govindachari, S. Rajappa and V. Sundarasanam, Indian J. Chem., 1963, 1, 247.

10. C.W. Moser, O.P. Crews, E.M. Acton and L. Goodman, J. Medicinal. Chem., 1966, 9, (2), 237.

11. R.N. Stillwell, Ph.D Thesis, Harvard University, 1964 Dissertation Abstracts, 1964, 64 - 11, 563.

12. L.K. Dalton, S. Demerac, B.C. Elmes, J.W. Loder, J.M. Swan and T. Teitei, Aust. J. Chem., 1967, 20, 2715.

13. B.C. Elmes, and J.M. Swan, Aust. J. Chem., 1969, 22, 1963.

14. A. Wander, Swiss patent 386, 023 (C.I.C07d) (1966).

15. F. Le Goffic, A. Gouyette and A. Ahond, C.R. Acad. Sci. Ser. C., 1972, 274, 2008 and Tetrahedron, 1973, 29, 3357.

16. K.N. Kilminster and M. Sainsbury, J. Chem. Soc., Perkin I, 1972, 2264.

17. B. Webb and M. Sainsbury, J. Chem. Soc., Perkin I, 1974, 1580.

18. M. Sainsbury, B. Webb and R.F. Schinazi, J. Chem. Soc., Perkin I, 1975, 289.

19. D. Dolman, M.Sc. Thesis, University of Bath, 1978 122 and 144.

20. Y. Tamura, J. Minamikawa, Y. Miki, S. Matsugashita and M. Ikeda, Tetrahedron Letters; 1972, 40, 4133.

21. Y. Tanura, J. Minamikawa and M. Ikeda, Synthesis, 1977, 1, 1.

22. J.G. Moffat, Techniques and Applications in Organic Synthesis, Oxidation, Marcel and Decker, New York, Ed. R.C. Augustine and D. Trecher, 1971, Vol. 2 p 1-59, and references therein.

23. S.N. Rastogi, J.S. Bindra, S.N. Rai and N. Anand, Indian J. Chem., 1972, 10, (6), 673.

24. J. Schmutz and H. Wittwer, Helv. Chim. Acta., 1960, 43, 793.

25. E. Wenkert, and D.G. Dave, J. Org. Chem., 1962, 45, 94.

26. M.J. Winchester and F.D. Popp, J. Heterocyclic Chem., 1975, 547.

27. J.P. Kutney and D.S. Grierson, Heterocycles, 1975, 3, 171.

28. Y. Oikawa and O. Yonemitsu, J. Chem. Soc., Perkin I, 1976, 1479.

29. (a) R. Besselievre, C. Thal, H.P. Husson and P. Potier, J. Chem. Soc., Chem. Comm, 1975, 90.

(b) Y. Langlois, N. Langlois and P. Poitier, Tetrahedron Letters, 1975, 955.

30. (a) T. Kametani, Y. Ichikawa, T. Suzuki and K. Fukumoto, Heterocycles, 1975, 3 (5), 401.

(b) T. Kametani, Y. Ichikawa, T. Suzuki and K. Fukumoto, Heterocycles, 1974, 2 (2), 171, and J. Chem. Soc., Perkin I, 1975, 413

31. R. Besselievre and H.P. Husson, Tetrahedron Letters, 1976 1873.

32. S.J. Martinz and J.A. Joule, J. Chem. Soc., Chem. Comm, 1976, 818.

33. J.A. Joule and D.A. Taylor, J. Chem. Research, Synopses, 1979, (12), 387.

34. V. Snieckus, Heterocycles, 1980, 14 (10), 1672.

35. C. Rivalle, C. Ducrocq and E. Bisagni, J. Chem. Soc. Perkin Trans I., 1979, 138.

36. F. Nivoliers, A. Decormeille, A. Godard and G. Queguiner, Tetrahedron Letters, 1980, 21, 4485.

37. W.L.F. Armarego, J. Chem. Soc., 1962, 1, 4094.

38. E.M. Hawes and D.K.J. Gorecki, J. Heterocyclic Chem., 1974, 11, 151 and references cited therein.

39. E.M. Hawes and D.K.J. Gorecki, J. Heterocyclic Chem., 1972, 9, 703.

40. R.B. Miller and T. Mook, Tetrahedron Letters, 1980, 21, 3319.

41. H.O. House, Modern Synthetic Reactions, Benjamin Incorp. New York, 2nd Edition 1972, p. 358.

42. (a) I. Goldberg, Chem. Ber., 1907, 40, 4541.

(b) P.E. Weston and H. Adkins, J. Amer. Chem. Soc., 1959, 50, 859.

(c) A.H. Jackson, P.R. Jenkins, and P.V.R. Shannon, J. Chem. Soc., Perkin I, 1977, 1698.

43. B. Akermark, L. Eberson, E. Jonsson and E. Pettersson, J. Org. Chem., 1965, 40, 1365.

44. W. Carruthers, J. Chem. Soc. Chem. Comm., 1966, 272 and K. Grellmann, G. Sherman and H. Linschitz, J. Amer. Chem. Soc., 1963, 85, 1881.

45. J. Bergman and R. Carlsson, Tetrahedron Letters, 1977, 4663.

46. A.P. Kozikowski and N.M. Hasan, J Org. Chem., 1977, 42 (11), 2039.

47. E.E. Harris, R.A. Firestone, K.Pfister, R.R. Boettcher, F.J. Cross, R.B. Currie, M. Monaco, E.R. Peterson and W. Reuter, J. Org. Chem., 1962, 27, 2705; R.A. Firestone, E.E. Harris, and W. Reuter, Tetrahedron, 1967, 23, 943; P.F. Muhlradt, Y. Marino, and E.E. Snell, J. Med. Chem., 1967, 10, 341.

48. H.B. Henbest, E.R.H. Jones, and G.F. Smith, J. Chem. Soc., 1953, 3796.

49. V.S. Rozhkov, Y.I. Smushkevich and N N. Suvorov, J. Heterocyclic Chem , 1976, 11, 826.

50. P.J. Mill and W.R.C. Crimmin, Biochim. Biophys. Acta, 1957, 23, 432; N.N. Suvorov, B. Ya. Eryshew, L.E. Frumin and A.G. Dubinin, J. Heterocyclic Chem., 1976, 10, 1325.

51. U. Schöllkopf and R. Schröder, Angew Chem., Int. Ed. Engl., 1971, 10, 333.

52. J.A. Joule and G.F. Smith, Heterocyclic Chemistry, Van Nostrand Reinhold Co. London, 1972, p. 315.

53. M. Sainsbury and R.F. Schinazi, J. Chem. Soc., Chem. Comm., 1975, 540.

54. M. Sainsbury and R.F. Schinazi, J. Chem. Soc., Perkin Trans I, 1976, 1155.

55. J.I. De Graw, J.G. Kennedy and W.A. Skinner, J. Heterocyclic Chem., 1966, 3, 67.

56. J.Y. Lallemand, P. Lemaitre, L. Beeley, P. Lesca, and D. Mansuy, Tetrahedron Letters, 1978, 15, 2161.

57. M.N. Dat-Xuong, M.T. Adeline, M.P. Lecointe and M.M. Janot, C.R. Acad. Sci. Paris, 1975, Series C 281 (15), 623.

58. V.N. Reinhold, L. Bittman, R. Bruni, K. Thrun and D. Silveira, Proc. Am. Assoc. Cancer Research, 1975, 16, 135

59. V.N. Reinhold and R.J. Bruni, Biomedical Mass Spectrometry, 1976, 3, 335.

60. A. Kalir, Israel. J. Chem., 1967, 5, 129.

61. M. Julia and J.Y. Lallemand, Bull. Soc. Chim. France, 1973, 2046.

62. N. Van-Bac, M. Herbert, L. Pichat, M.M. Janot, and N. Dat-Xuong, J. of Labelled Comp., 1974, 11 (2), 241.

63. M.M. Chien and J.P. Rosazza, Drug Metabolism and Disposition, 1979, 7 (4), 211.

64. R.E. Hartman, E.F. Kraus, W.W Andres, and E.L. Patterson, Applied Microbiology, 1964, 12, 138.

65. D.R. Brannon, D.R. Horton and G.H. Svoboda, J. Med. Chem., 1974, 17, 653.

66. R.V. Smith and J.P. Rosazza, J. Pharm. Sci., 1975, 64, 1737.

67. M. Luckner, Secondary Metabolism in Plants and Animals, Chapman and Hall, London, 1972, 43.

68. G. Mathe, M. Hayat, F. de Vassal, L. Schwarzenberg, M. Schneider, J.R. Schlumberger, C Jasmin and C. Rosenfield, Rev. Europ. Etud. Clin. Biol., 1970, 15, 541.

69. M. Driver, I.T. Mathews and M. Sainsbury, J. Chem. Soc., Perkin I, 1979, 10, 2506.

70. J.F.W. McOmie, Review of Protective groups, Advances in Organic Chemistry: Methods and Results, 1963, 3, 191.

71. T. Schmidt and A. Schach, Ann., 1951, 29, 571.

72. J. Davoll and D.H. Laney, J. Chem. Soc., 1956, 2124.

73. R.J. Sundberg, The Chemistry of Indoles, Academic Press, New York, 1970, p.142.

74. F.T. Tyson, Organic Syntheses; Collective Volume III; Edited by E.C. Horning, John Wiley & Sons Incorp. 1964, 479.

75. J.A. Joule and G.F. Smith, Heterocyclic Chemistry, Van Nostrand Reinhold, London 1972, p. 285.

76. R.M. Acheson, An Introduction to the Chemistry of Heterocyclic Compounds, Second Edition, John Wiley & Sons Ltd., New York, 1967, p. 168.

77. R.A. Heacock, O. Hutzinger, B.D. Scott, J.W. Daly and B. Witkop, J. Amer. Chem. Soc., 1963, 85, 1825.

78. A. Eck and B. Witkop, J. Amer. Chem. Soc., 1954, 76, 5579.

79. B. Robinson, Chemical Reviews, 1963, 63, 373.

80. J.T. Fitzpatrick and R.D. Hiser, J. Org. Chem., 1957, 22, 1703.

81. T.L. Gilchrist and R.C. Storr, Organic reactions and Orbital Symmetry, Cambridge University Press, 1972, p. 234.

82. B. Marchand, C. Streffer, and J. Jauer, J. Prakt Chem., 1961, 13, 54.

83. K.G. Blaike and W.H. Perkin, J. Chem. Soc., 1924, 125, 296.

84. T.R. Govindachari, P. Madhaven Pillai, K. Nagarajan and N. Viswanathan, Tetrahedron, 1965, 21, 2957.

85. N.N. Suvorov, M.V. Fedotova, L.M. Orlova and O.B. Ogareva, Zn. Obshch Khim., 1962, 32, 2358 and Chem. Abstract, 1963, 58, 9007.

86. R.L. Hinman and J. Lang, J. Amer. Chem. Soc., 1964, 86, 3796, and papers cited therein.

87. J.D. Benigni and R.L. Minnis, J. Heterocyclic Chemistry, 1965, 35, 387.

88. T.F. Spande and W.A. Remers, The Chemistry of Heterocyclic Compounds, Indoles Part 3, Ed. W.J. Houlihan, John Wiley and Sons, New York, 1979, p. 25.

89. H. Burton and J.L. Stoves, J. Chem. Soc., 1937, Part II, 1726.

90. R.J.S. Beer, K. Clarke, H.G. Khorana and A. Roberton, J. Chem. Soc., 1948, Part II, 1605.

91. W.E. Noland and F.J. Baude, Organic Syntheses, Collective Volume V, Edited by Baumgarten, John Wiley and Sons Incorp., 1973, p. 567.

92. M. Copisarow, J. Chem. Soc., 1929, Part I, 251.

93. Prepared by a modification of the procedure employed by L.F. Fieser and S. Rajagoplan, J. Amer. Chem. Soc., 1949, 71, 3938.

94. F.A. Mason, J. Chem. Soc., 1925, 1, 1195.

95. A. Lovecy, R. Robinson and S. Sugasawa, J. Chem. Soc., 1930, 817.

96. See reference 75, p. 287.

97. H.N. Rydon and J.C. Tweddle, J. Chem. Soc., 1955, Part III, 3499.

98. F. Bergel and A.L. Morrison, J. Chem. Soc., 1943, 49.

99. W.O. Kermack, W.H. Perkin and R. Robinson, J. Chem. Soc., 1922, 121, 1872.

100. M. Node, H. Hori and E. Fujita, J. Chem. Soc., Perkin I, 1976, 2237.

101. G. Buchi, D. Spitzner, S. Paglialunga and G.N. Wogan, Life Sciences, 1973, 13, 1143.

102. C.F. Huebner, H.A. Troxell and D.C. Schroeder, J. Amer. Chem. Soc., 1953, 75, 5887.

103. T.K. Kralt, W.J. Asma, H.H. Haeck, and H.D. Moed, Rec. Trav. Chim., 1961, 80, 313.

104. Based on a method for the preparation of meta-nitroaniline, from A.I. Vogel, Practical Organic Chemistry, Longman Green & Co. Ltd., London, Third Edition, 1964, p. 574. Also V. Cunerth, Annallen, 1874, 172, 228. For preparation of 4-methyl-3-nitroaniline.

105. Based on a method for the preparation of meta-nitrophenol, from A.I. Vogel, Practical Organic Chemistry, Longman

Green & Co. Ltd., London, Third Edition, 1964, p. 614. Also Nevil and Winther Chem. Ber., 1882, 15, 2980. For preparation of 4-methyl-3-nitrophenol.

106. Based on a method for the preparation of anisidine by C.S. Hiers and F.D. Hayer, Organic Synthesis; Collective Volume I, Edited by A.H. Blatt and H.G. Gilman, John Wiley & Sons Incorp, London, Second edition 1964, p. 58.

107. W.R. Boon, J. Chem. Soc., 1940, S, 230.

108. Based on a method for the preparation of 4-nitrobenzaldiacetate and 4-nitrobenzaldehyde by S.V. Lieberman and R. Conner, Organic Synthesis; Collective Volume II, Edited by A.H. Blatt, John Wiley and Sons Incorp., 1943, p. 441.

109. R.B. Woodward, F.E. Bader, H. Bickel, A.J. Frey and R.W. Kierstead, Tetrahedron, 1958, 2, 1.

110. D.E. Worrall, Organic Syntheses; Collective Volume I, Second Edition, 1964, p. 413.

111. W.E. Noland, L.R. Smith and K.R. Rush, J. Org. Chem., 1965, 30, 3457, and references cited therein.

112. L. Marion and C.W. Oldfield, Can. J. Res., 1947, 25B, 1.

113. T. Sugasawa, M. Adachi, K. Sasakura, and A. Kitagawa, J. Org. Chem., 1979, 44, 578.

114. H.G. Kolloff and J.H. Hunter, J. Amer. Chem. Soc., 1941, 63, 490.

115. R.T. Morrison and R.N. Boyd, Organic Chemistry, Allyn and Bacon Incorp., Boston, Second Edition, 1972, p. 399.

116. W.R. Reppe, O. Schlichting, K. Klager and T. Toepel, Ann., 1948, 560, 1.

117. H.A. Iddles, E.H. Lang and D.C. Gregg, J. Amer. Chem. Soc., 1937, 2, 1945.

118. Unpublished communication from Professor J. Bergman, Royal Institute of Technology, Stockholm, Sweden.

119. See reference 73, p. 38.

120. See reference 75, p. 273.

121. M. Driver, Ph.D Thesis, University of Bath, p. 77.

122. C.H. Depuy and O.L. Chapman, Molecular Reactions and Photochemistry, Ed. K.L. Rinehart, Prentice Hall Incorp., Englewood Cliffs, New Jersey, 1972, p. 37.

123. A.N. Terenin and V.A Ermolaev, Trans. Faraday Soc., 1956, 52, 1042.

124. J.K. Kochi and G.S. Hammond, J. Amer. Chem. Soc., 1953, 75, 3443.

125. D. Watkins, Ph.D Thesis, University of Bath, 1982, p. 64.

126. W. McCrae, Basic Organic Reactions, Heyden and Son Ltd., New York, 1973, p. 59.

127. L.F. Feiser and M. Fieser, Reagents for Organic Synthesis, John Wiley & Sons Incorp., Volume I, 1967, 1210.

128. M. Hooper and W.N. Pitkethly, Journal of the Chemical Society, Perkin I, 1973, 2804.

129. M. Hooper and W.N. Pitkethly, Journal of the Chemical Society, Perkin I, 1972, 1607.

130. A. Lewis and J. Meinwald, J. Amer. Chem. Soc., 1961, 83, 2769.

131. D.C. Neckers, Mechanistic Organic Photochemistry, Reinnold, New York, 1967, p. 116.

132. P.D. Bartlett, Science, 1968, 159, 833

133. P.D. Bartlett, Pure and Applied Chemistry, 1971, 27, 597.

134. H.M. Frey and R.M. Walsh, Chemical Reviews, 1969, 69, 103.

135. A.J. Cocks, H.M. Frey and I.D.R. Stevens, J. Chem. Soc., Chem. Comm., 1969, 458.

136. J.E. Baldwin and P.W. Ford, J. Amer. Chem. Soc., 1969, 91, 7192.

137. R. Grigg, Ann. Reports, 1971, 68B, 154.

138. R.B. Woodward and R. Hoffmann, Angew. Chem. Internat. Edn., 1969, 8, 781.

139. W.N. Pitkethly, Ph.D Thesis, C.N.A.A., 1972, pp 83 and 98.

140. Prepared by the procedure for methyltriphenylphosphonium bromide used by I.T. Harrison, B. Lythgoe and S. Trippett, J. Chem. Soc., 1955, Part 4, 4016.

141. W.S. Wadsworth, Organic Reactions, (Synthetic applications of phosphoryl stabilized anions), 1977, 25, 73.

142. G.P. Schiemenz and J. Thobe, Chem. Ber., 1966, 99, 2663.

143. M. Schlosser and K.F. Christmann, Angew. Chem. Intern. Edn., 1964, 3, 636.

144. H.O. House, Modern Synthetic Reactions, Benjamin Incorp., New York, 1965, p. 690.

145. W.S. Wadsworth and W.D. Emmons , J. Amer. Chem. Soc., 1961, 83, 1733.

146. H.O. House and M. Schellenbaum, J. Org. Chem., 1963, 28, 34.

147. K. Kilminster, Ph.D Thesis, University of Bath, 1972, p. 103.

148. M. Driver, Ph.D Thesis, University of Bath, 1979, p.20.

149. P.L. Julian and A. Magnani, J. Amer. Chem. Soc., 1949, 71, 3207.

150. Brunner and Moser, Monatsch, 1932, 61, 15.

151. G. Tacconi, S. Pietra and M. Zaglio, Farmaco. Ed. Sci., 1965, 20, 470.

152. E. Wenkert, B.S. Berstein and J.H. Udelhofen, J. Amer. Chem. Soc., 1958, 80, 4899.

153. M. Julia, F. Le Foffie, J. Igolen and M. Baillarge, Tetrahedron Letters, 1969, 20, 1569.

154. H. Sirowej, S.A. Khan and H. Plieninger, Synthesis, 1972, 2, 84.

155. N. Aimi, E. Yamanaka, J. Endo, S. Sakai and J. Haginiwa, Tetrahedron Letters, 1972, 11, 1081.

156. N. Umino, T. Iwakuma and N. Itoh, Tetrahedron Letters, 1976, 10, 763.

157. H. Pleininger and G. Werst, Angew. Chem. Intern. Edn., 1958, 70, 272.

158. A. Kubo and T. Naki, Synthesis, 1980, 5, 365.

159. Prepared by a modification of the procedure for phenylacetic acid by A.I. Vogel, Practical Organic Chemistry, Longmans, Green & Co. Ltd., London, Third Edition 1964, p. 758. Also Borsche, Ber., 1909, 42, 1313.

160. R. Adams and A.F. Thal, Organic Syntheses; Collective Volume I, Second Edition Ed. H.G. Gilman and A.H. Blatt, 1941, 436.

161. See reference 83, p. 308.

162. E. McDonald and R.D. Wylie, Tetrahedron, 1979, 35, 1415.

163. See reference 75, p. 278.

164. A.H. Beckett, R.W. Daisley and J. Walker, Tetrahedron, 1968, 24, 6093.

165. F.W. Neumann, N.B. Sommer, C.E. Kaslow, and R.L. Shriner, Organic Syntheses; Collective Volume III, 1955, 519.

166. M. Freifelder, J. Org. Chem., 1966, 31, 3875.

167. L. Horner, H. Reuter, and E. Herrmann, Justus Liebigs Ann. Chem., 1962, 1, 660.

168. H. Adkins and R.E. Burks, J. Amer. Chem. Soc., 1948, 70, 4174.

169. See reference 73, p. 37.

170. R.W. Guthrie, A. Brossi, F.A. Mennona, J.G. Mullin, R.W. Kierstead and E. Grunberg, Journal of Medicinal Chemistry, 1975, Volume 18, No. 7,755.

171. A.H. Jackson and G.W. Stewart, J. Chem. Soc., Chem. Comm., 1971, 149.

172. Jan. Bergman, R. Lars and B. Sjöberg, Tetrahedron, 1980, 36(17), 2505-11.

173. B. Sjöberg, Ph.D Thesis, Royal Institute of Technology, Stockholm, 1979, Part II, p. 16.

174. I. Matthews, Ph.D Thesis, University of Bath, 1978, p. 84.

175. I.L. Finar, Organic Chemistry, Volume I, Fifth Edition, Longmans Green and Co. Ltd., London, 1967, pp. 616, 670 and 762.

176. Unpublished work by Dr. B. Webb, University of Bath, 1974.

177. Personal communication from Dr. B. Sjöberg, University of Bath, 1980.

178. R.O.C. Norman, Principles of Organic Synthesis, Methuen & Co. Ltd., 1968, p. 579.

179. See reference 7, p. 92.

180. W.C. Sumpter and F.M. Miller, The Chemistry of Heterocyclic Compounds, Interscience Publishers Incorp. New York, 1954, (Carbazoles) p. 70.

181. Mizuch and Sanchenko, J. Gen. Chem. (U.S.S.R.) 1940, 10, 852 - 854, and Chem. Abstr., 1941, 35, 2509.

182. H. Lindemann and F. Muhlhaus, Chem. Ber., 1925, B58, 2371.

183. N.G.P.H.Buu-Hoi, Rec. trav. chim., 1947, 66, 533.

184. Cosgrove and Waters, J. Chem. Soc., 1949, 907.

185. H.C.Longuet-Higgins and C.A. Coulson, Trans. Faraday Soc., 1947, 43, 87.

186. Personal communication from Dr. B. Astin, University of Bath, 1980.

187. H. Lindemann, Chem. Ber., 1924, 57, 555.

188. O. Ruff and V. Stein, Chem. Ber., 1901, 34, 1668.

189. Tucker and Stevens, J. Chem. Soc., 1923, 123, 2140, Kehrmann and Zweifel, Helv. Chem. Acta, 1938, 11, 1213, H. Furst and J. Bosse, Chem. Ber., 1951, 84, 83.

190. Ziersch, Chem. Ber., 1909, 42, 3800.

191. C. Gansser, C. Viel, C. Malry and S. Cros, Il Farmaco-Ed Sc., 1980, 35(11), 888.

192. A.R. Katritzky, H. Beltrami and M.P. Sammes, Journal of the Chemical Society, Perkin I, 1980 2480.

193. A.R. Katritzky, J.G. Keay, D.N. Rogers, M.P. Sammes and C.W.F. Leung, Journal of the Chemical Society, Perkin I, 1981, 588.

194. A.R. Katritzky, J.G. Keay and M.P. Sammes, Journal of the Chemical Society, Perkin I, 1981, 688.

195. M.P. Sammes, C.W.F. Leung, C.F. Mak and A.R. Katritzky, Journal of the Chemical Society, Perkin I, 1981, 1585.

196. R.E. Lyle and G.J. Gauthier, Tetrahedron Letters, 1965, 4615.

197. C.W.F. Leung, M.P. Sammes, and A.R. Katritzky, Journal of the Chemical Society, Perkin I, 1979, 1698.

198. A. Ohsawa, M. Hirobe, and T. Okamoto, J. Pharm. Soc. Jpn., 1972, 92, 73.

199. N. Rakieten, D.A. Cooney and R.D. Davis, U.S. Govt. Res. Develop. Rep. 1967, 67, 38.

200. G.H. Svobda, G.A. Poore and M.L. Montford, J. Pharm. Sci., 1968, 57, (10), 1720.

201. J. Le Men, M. Hayat, G. Mathe, J.C. Guillon, E. Chenu, M. Humblot and Y. Masson, Rev. Euro. Etud. Clin. Biol., 1970, 15 (5), 534.

202. R.T. Hill and R. Baserga, Cancer Treatment Review, 1975, 2, 159.

203. B. Festy, J. Poisson and C. Paoletti, F.E.B.S., (Fed. Euro. Biochem. Soc.) Letts., 1971, 17 (2), 321.

204. W.B. Pratt, Fundamentals of Chemotherapy, Oxford University Press, 1973, 309.

205. R. O'Brien, J. Olenick and F. Hahn, Proc. Nat. Acad. Sci. U.S.A., 1966, 55, 1511.

206. H.E. Skipper, F.M. Schabel and W.S. Wilcox, Cancer Chemotherapy Reports, 1965, 45, 5.

207. M. Yudkin and R. Offord, Comprehensible biochemistry, Longman Group Ltd., 1973, 146.

208. L. Lerman, J. Mol. Biol., 1964, 10, 367.

209. J. Wang, J. Mol. Biol., 1974, 89, 783.

210. J.D. Watson, Molecular Biology of the Gene, W.A. Benjamin Inc., Menlo Park, California, 2nd Ed., 1970, 276.

211. J.N. Davidson, The Biochemistry of the Nucleic Acids, Chapman and Hall, London, 7th Ed., 1972, 145 and 166.

212. M. Waring, J. Mol. Biol., 1970, 54, 247.

213. A. Albert, Selective Toxicity (The Physiochemical Basis of Therapy), Chapman and Hall, London, 6th Ed., 1979, 360.

214. J.B. Le Pecq, N. Dat-Xuong, C. Gosse and C. Paoletti, Proc. Nat. Acad. Sci. U.S.A., 1974, 71, 5078.

215. K. Kohn, M. Waring, D. Glaubiger and C. Friedman, Cancer Research, 1975, 35, 71.

216. See reference 213, p. 193.

217. C. Hantsch, Cancer Chemotherapy Reports, Part I, 1972, 56 (4), 433.

218. D. Dolman and M. Sainsbury, Tetrahedron Letters, 1981, 22, 2119.

219. M. Sainsbury, D, Weerasinghe and D. Dolman, Journal of the Chemical Society, Perkin I; 1982, 2, 587.

220. Unpublished communication from C. Paoletti.

221. C. Paoletti, J.B. Le Pecq, N. Dat-Xuong, P. Lesca and P. Lecointe, Proc. Int. Congr. Chemother., (ed. W. Siegenthaler and R. Luethey), 1977, 2, 1195; and unpublished communication from N.Dat-Xuong.

222. M. Sainsbury, Synthesis, 1977, 7, 437.

223. P.C. Ferreira, G.B.M. Bettolo, and J. Schmutz, Experientia; 1959, 15, 179.

224. O. Neunnöffler and H.Kölbel, Ber., 1935, 68, 255.

225. R.A. Morton and A. McGookin, J. Chem. Soc., 1934, 909.

226. Limpach, Ber., 1891, 24, 4140.

227. H.H. Sisler, Inorganic Syntheses; 1946, Volume II, 205. ed. C. Fernelius, McGraw Hill and Co. Ltd., London.

228. J.M.L. Stephen, I.M. Tonkin and J. Walker, J. Chem. Soc., 1947, 1038.

229. Knecht, Ann., 1882, 215, 91.

230. A. Pictet and L. Duparc, Ber., 1887, 201, 3421.

231. H.N. Wingfield, W.R. Harlan and H.R. Hanmer, J. Amer. Chem. Soc., 1953, 75, 4364.

232. H. Meerwein, Organic Synthesis; Collective volume V 1973, 1080. Ed. F. Baumgarten, John Wiley and Sons, New York.

233. D. Raileanu, O. Constantinescu-Simon, E. Mosanu and C. Nenitzescu, Rev. Roum. Chim., 1967, 12 (2), 105.

234. G. Wittig and D. Wittenberg, Ann., 1957, 606, 1-23 and C.A. 1957, 52, 1970f.

235. Borshe, Ber., 1909, 42, 1313.

236. Gabriel and Meyer, Ber., 1881, 14, 824.

237. Holtmeyer, Zeitschrift für Chemie, 1867, 686, and Beil, 11 136.

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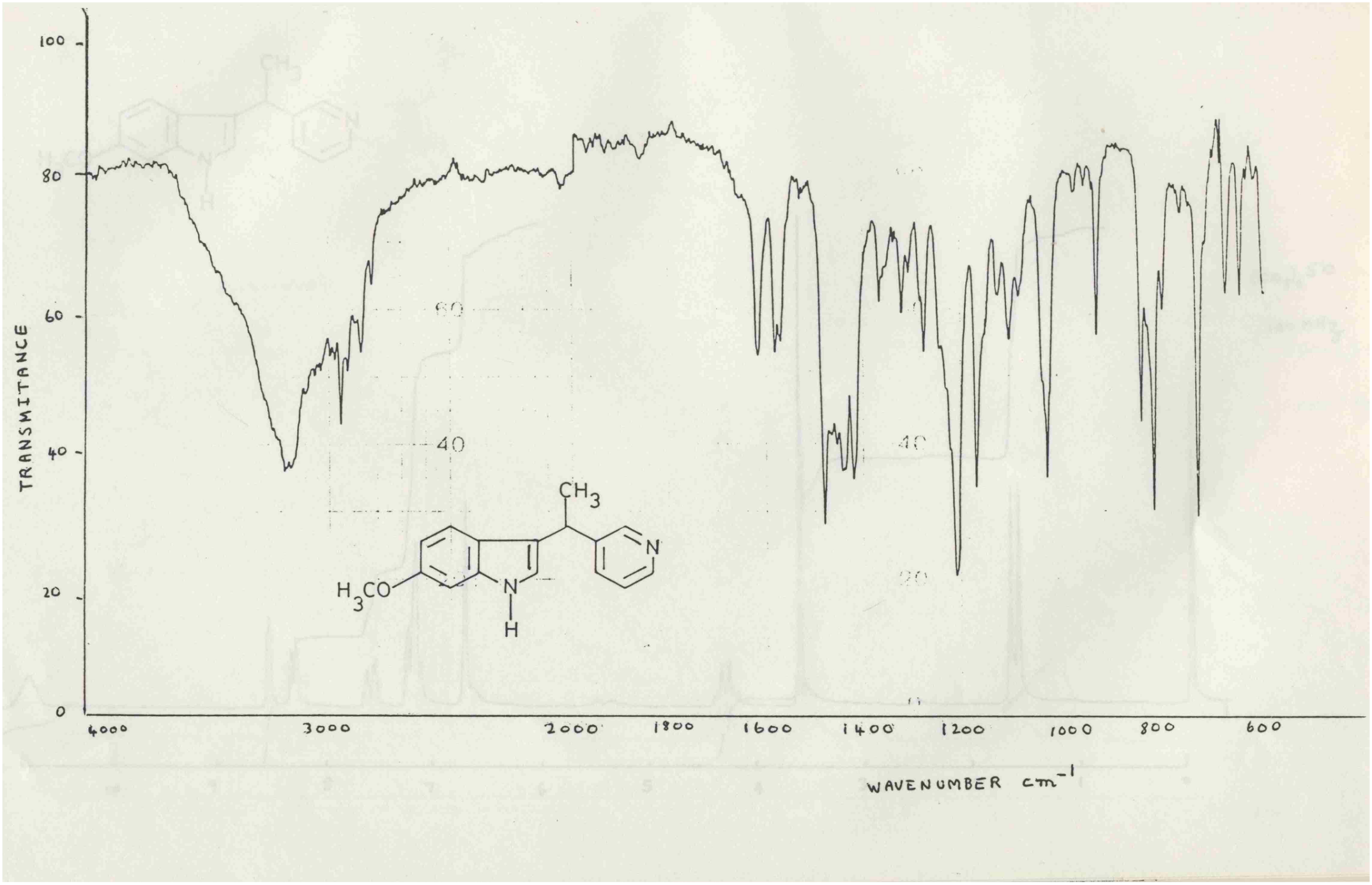
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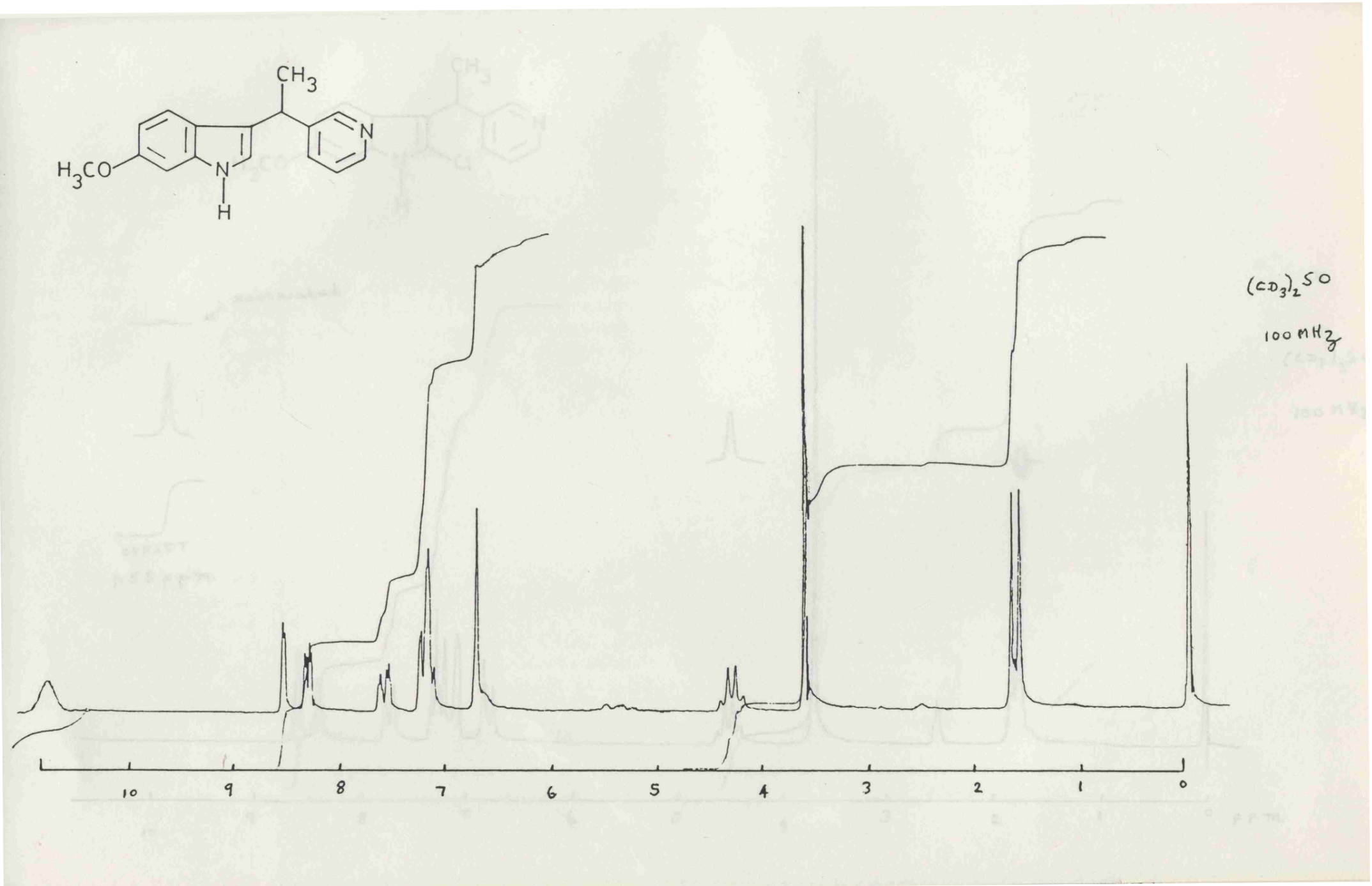
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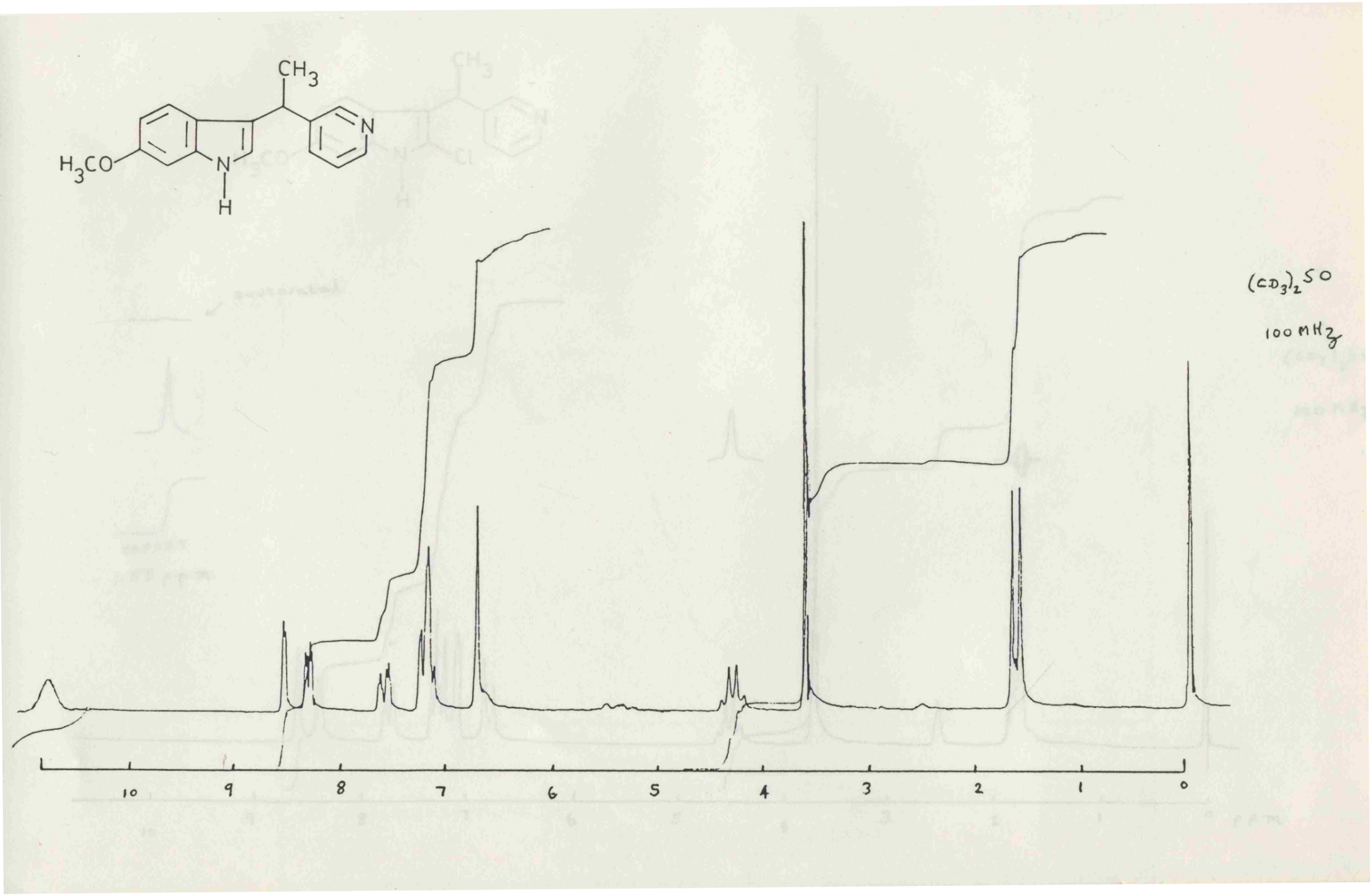
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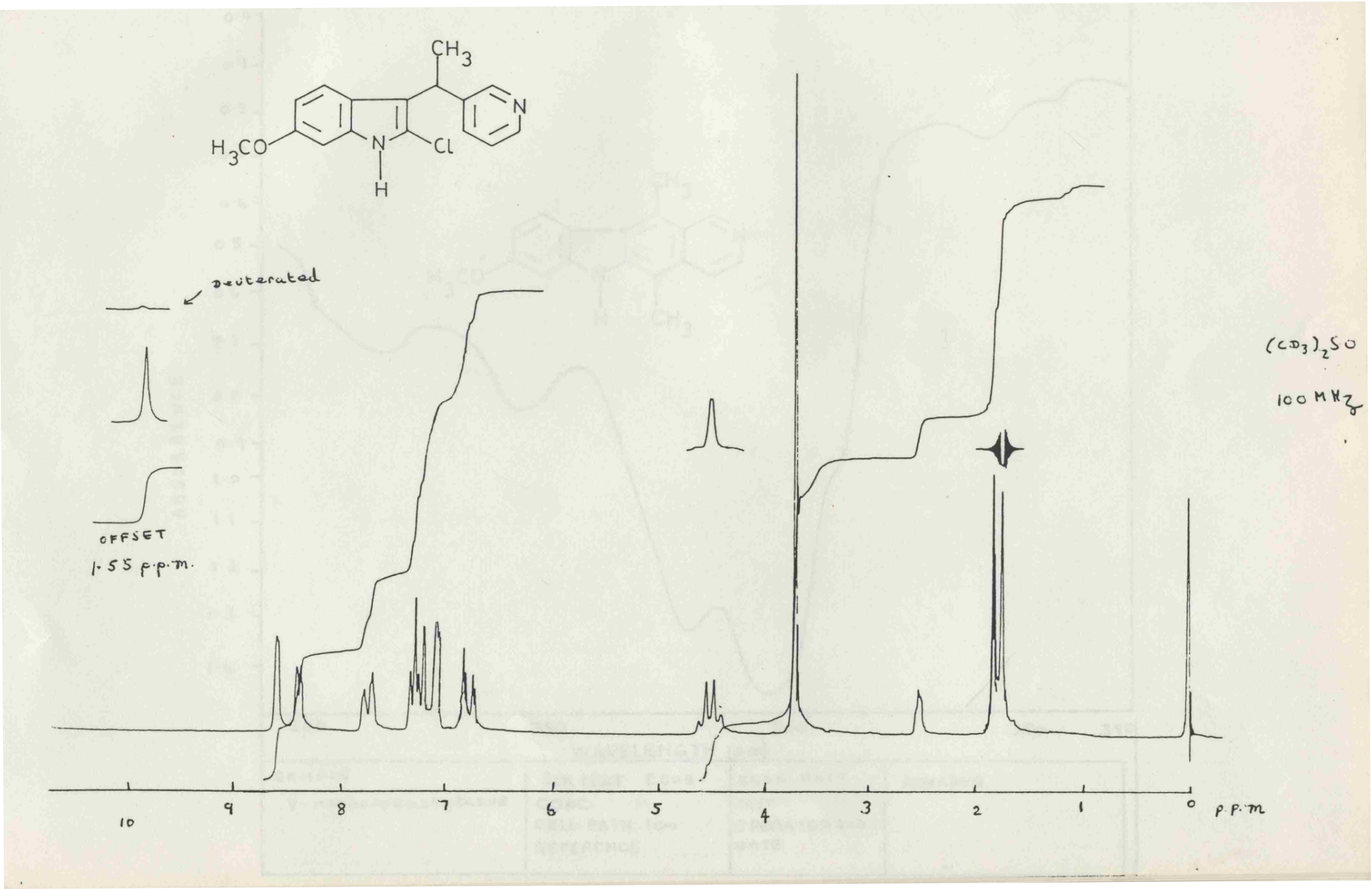
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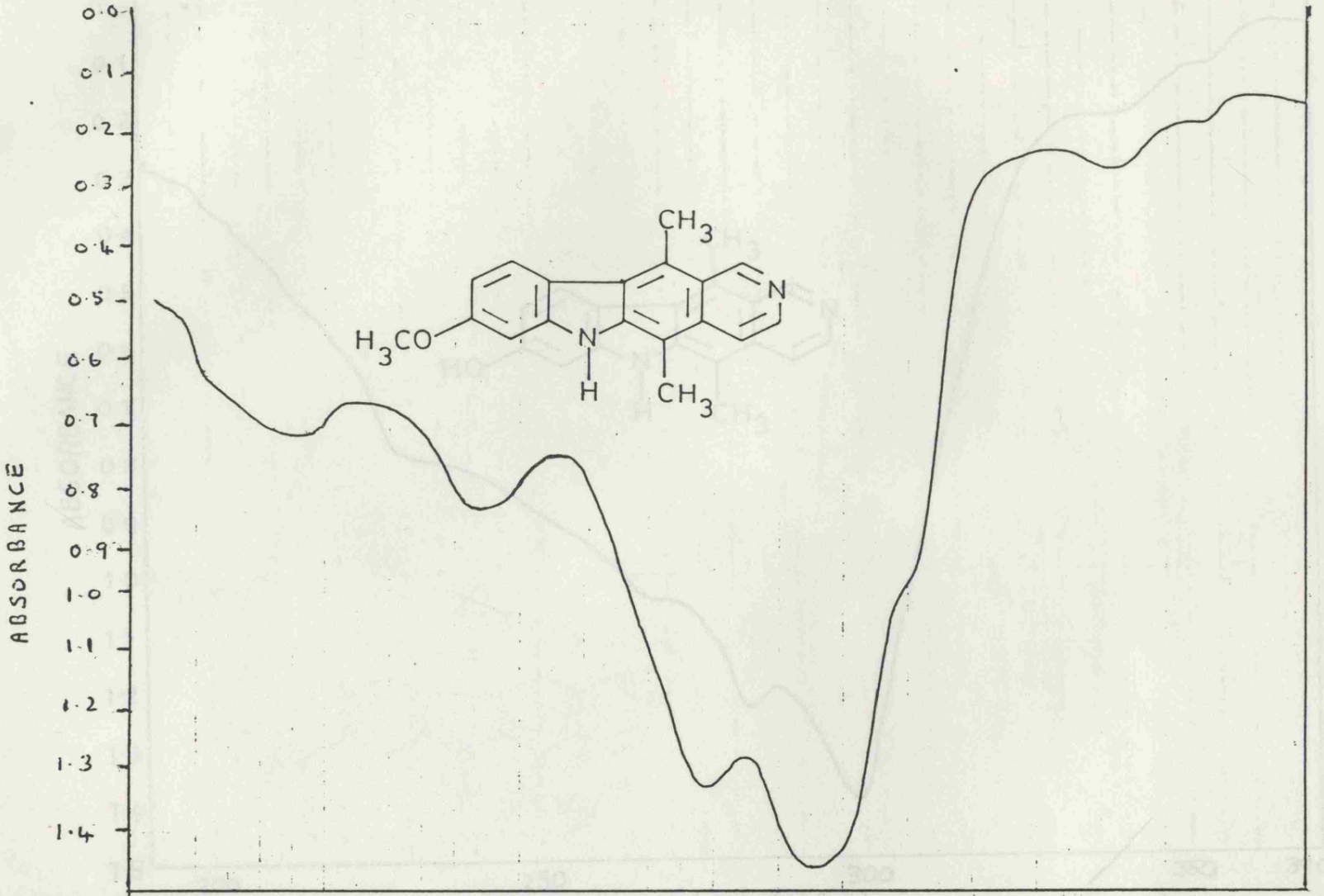

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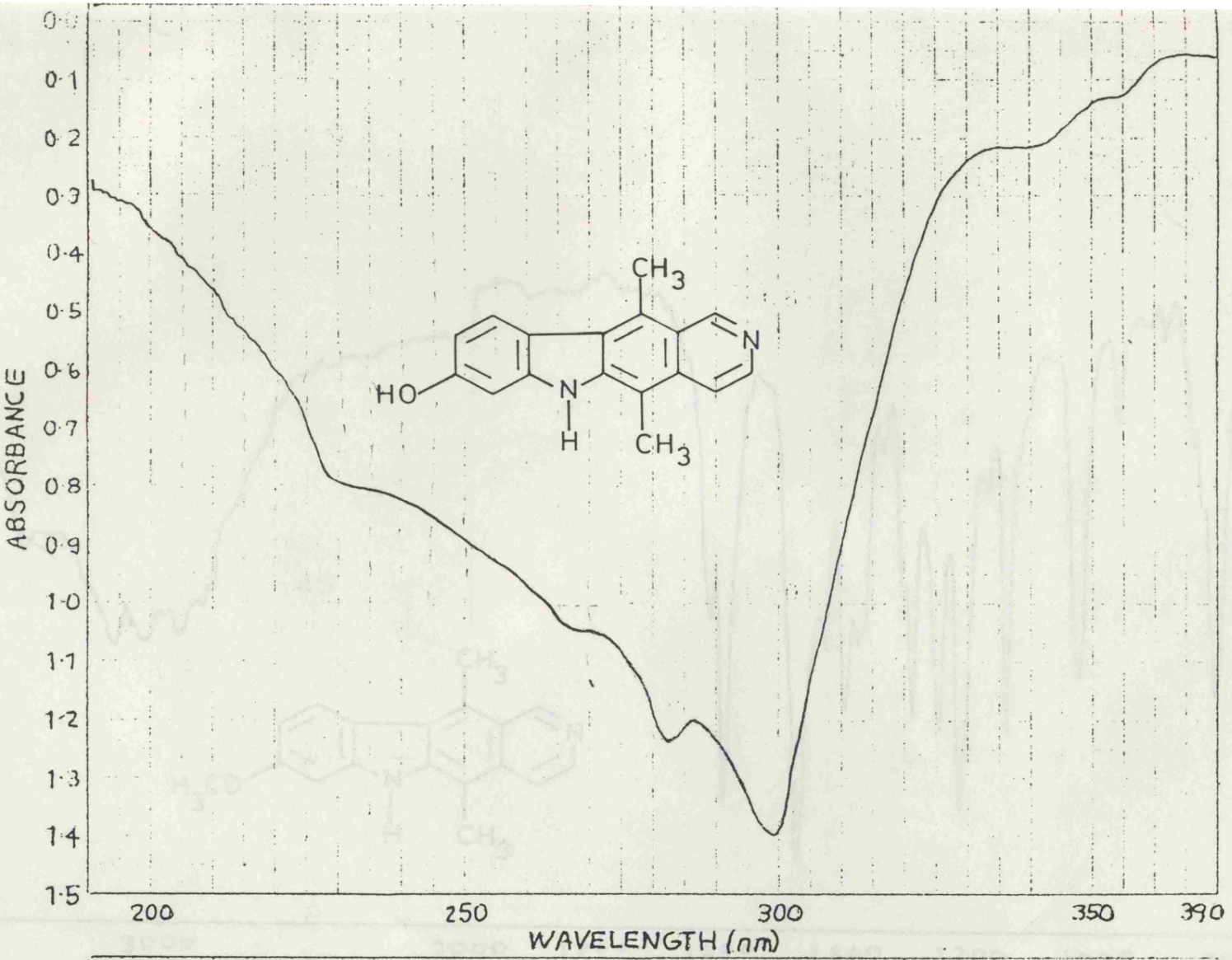
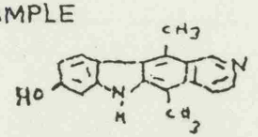


SAMPLE 	SOLVENT Ethanol	SCAN Fast	REMARKS
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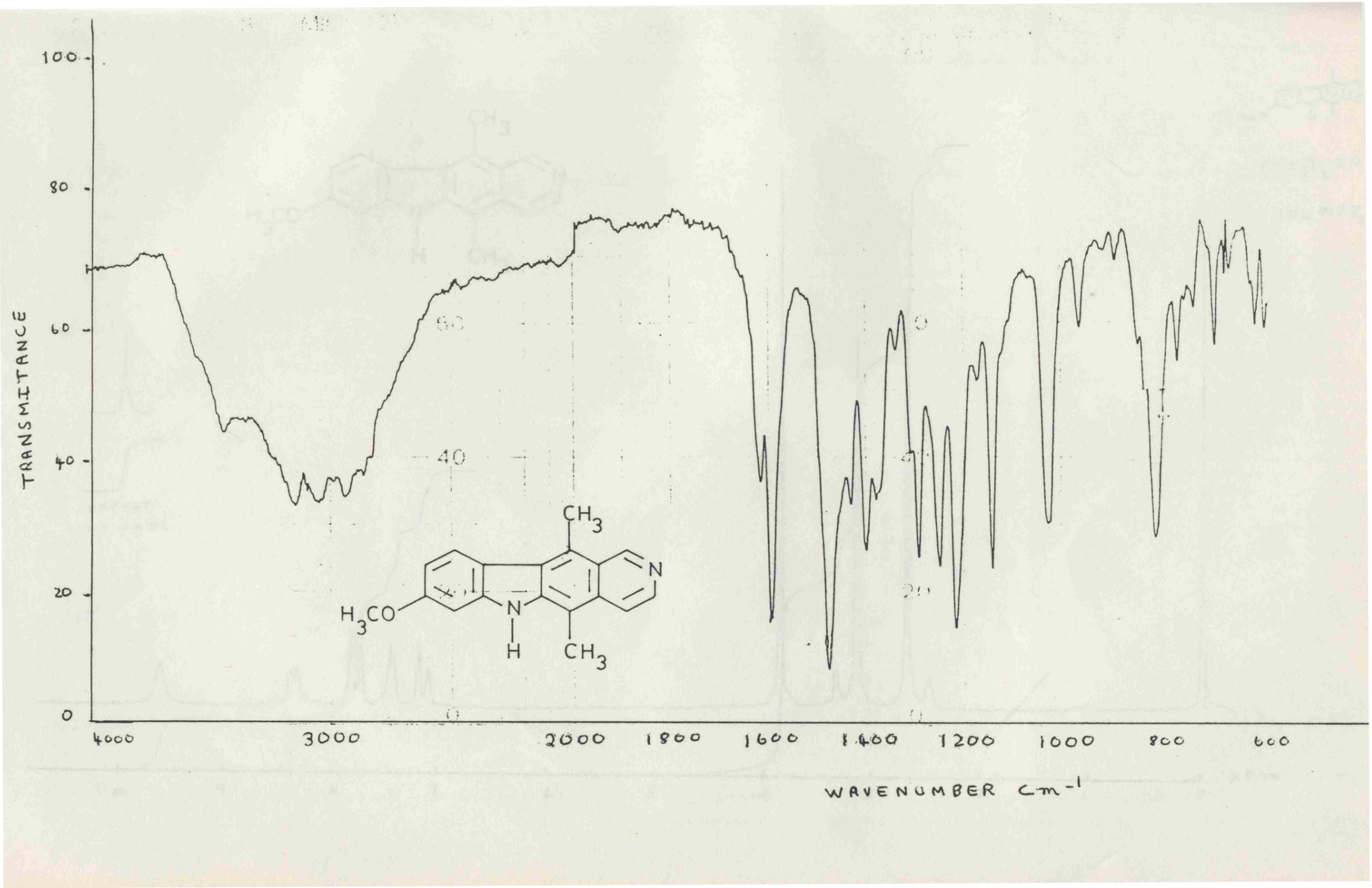
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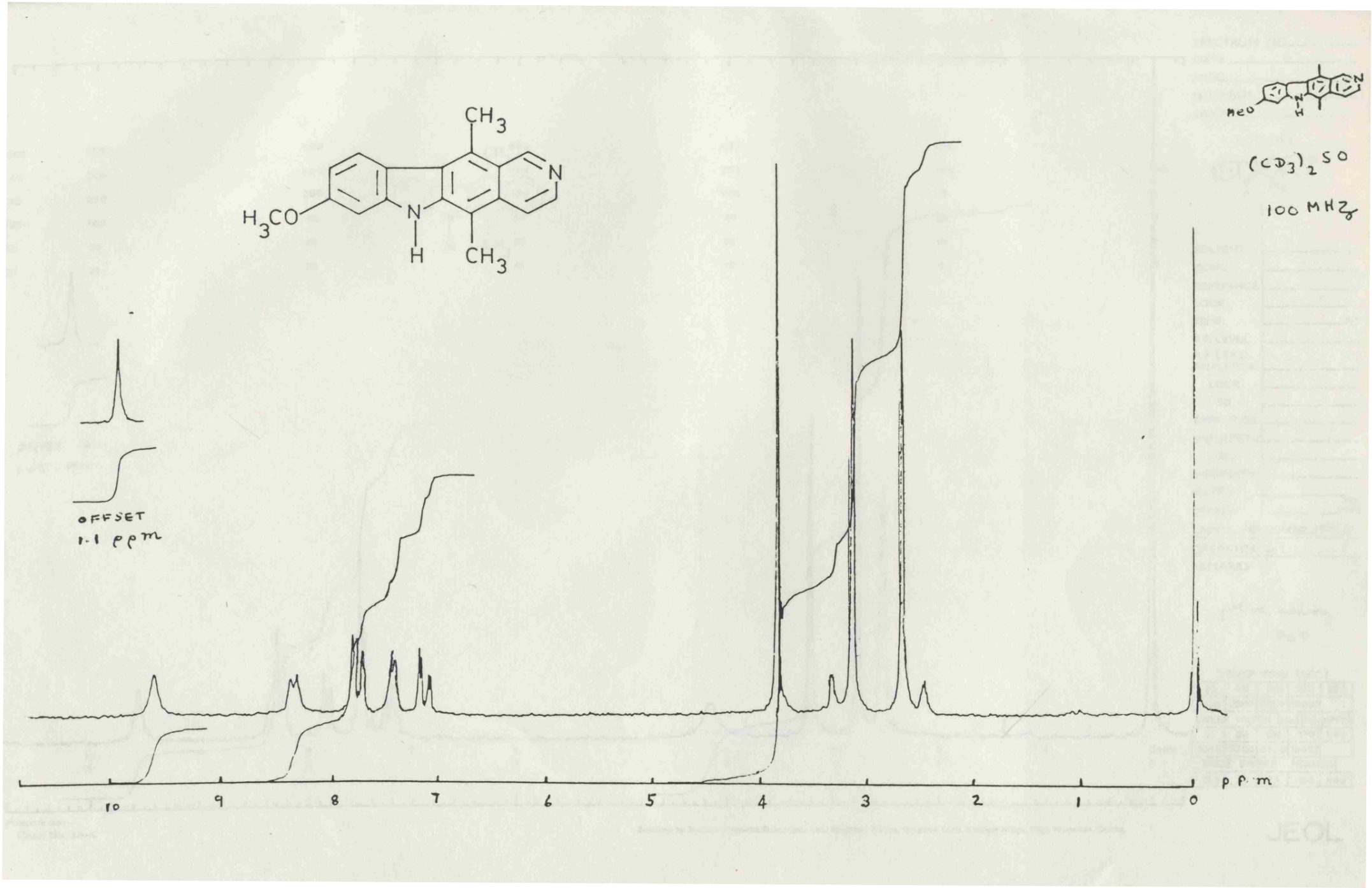
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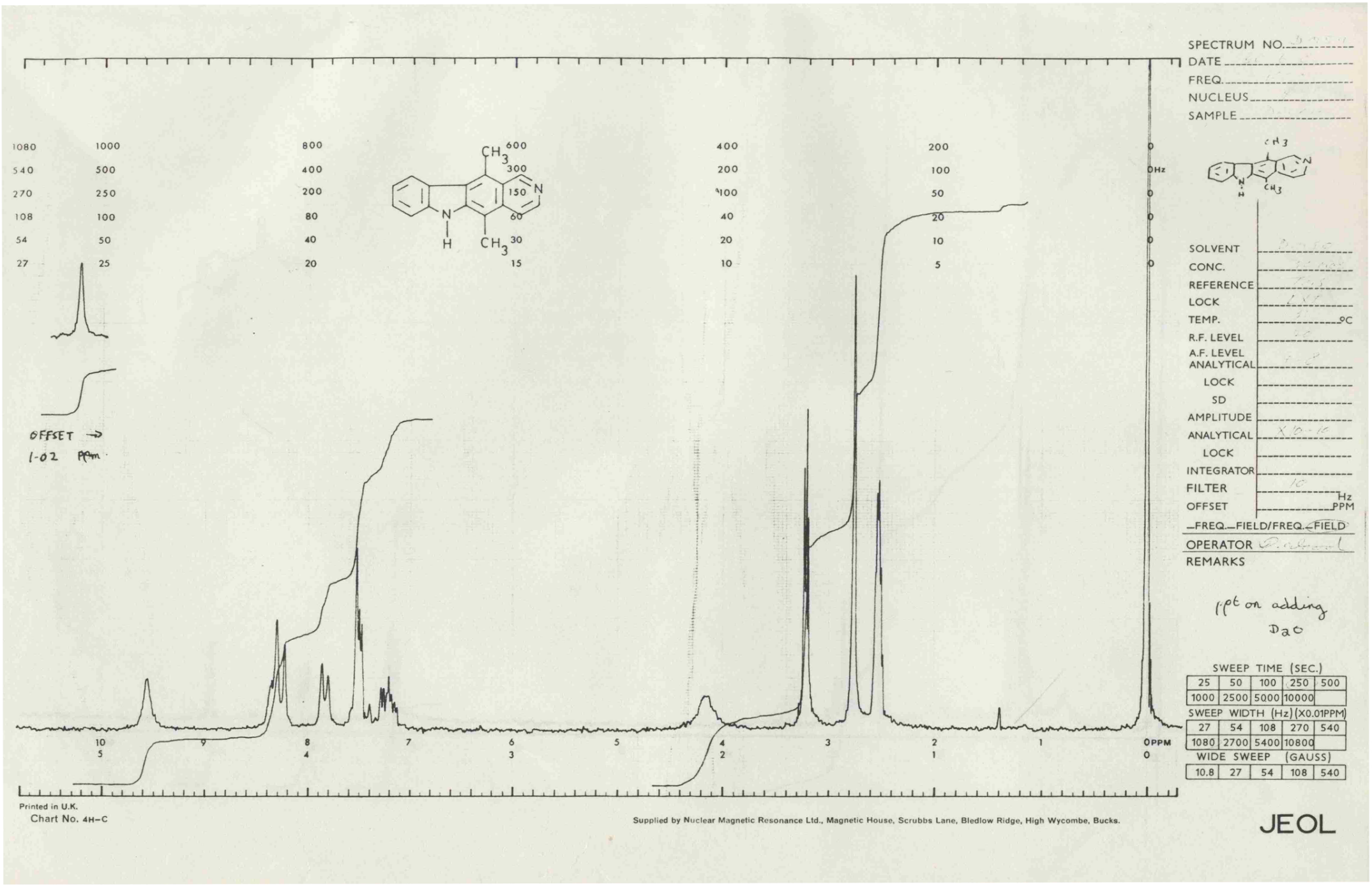
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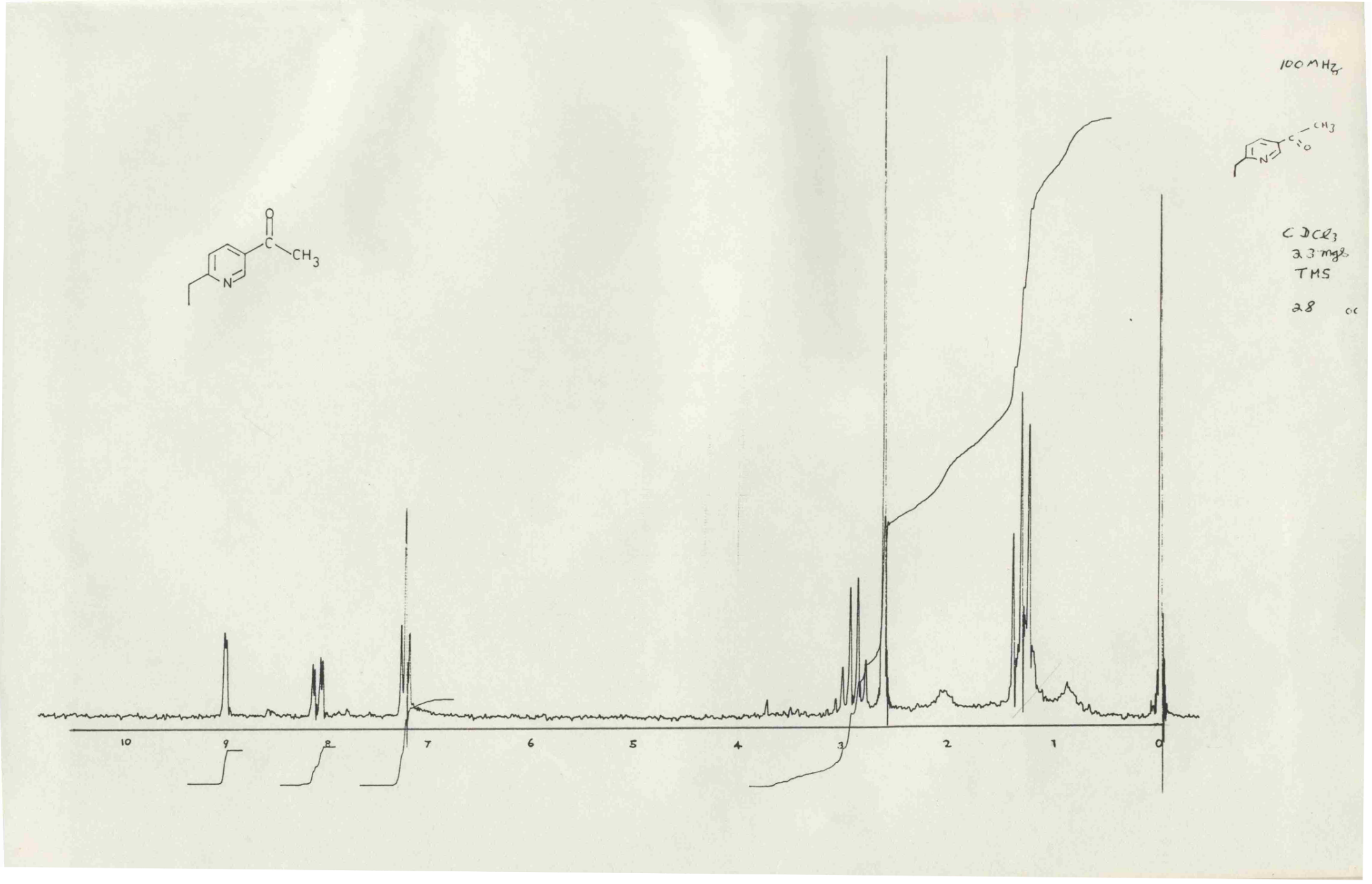
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